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ACIDOSIS DURING STARVATION

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INTRODUCTION

The physiological effects of starvation have been investigated by numerous observers and many papers have been published on the subject. Castellino noted a decrease in the alkalescence of the blood in starving rabbits in 1893, and in the following year Tanszk observed the same phenomenon in Succi's blood during a period of starvation. This result was confirmed by the careful studies of Benedict (1) in 1907. London also observed a slight decrease in blood alkalescence in starving rabbits. The appearance of acetone bodies in urine and expired air, which is characteristic of the late stages of starvation, has also been extensively studied. The output of carbon dioxide is said by various authors to show a steady reduction throughout periods of inanition. Thus Benedict reports that the carbon dioxide content of the alveolar air was subnormal on the second day of a fast. Thereafter it held constant until the fourteenth day, when a second drop occurred, after which it suffered no further reduction. In each case the fall in tension amounted to about 4 mm. Hg. Thus it may be inferred that a tendency toward acidosis developed in this individual on the second day and that this tendency toward acidosis was accentuated on the fourteenth day.

We know that the hydrogen ion concentration of the blood rather than the carbon dioxide tension is the predominating factor in the control of respiration, and also that the acidity of the blood may be divided into two parts, one due to carbon dioxide and another one

due to other acids. As the total hydrogen ion concentration necessary to stimulate the respiratory center must always be the same, it is readily seen that if the other acids in the blood increase in amount, the tension of carbon dioxide will decrease. Since alveolar carbon dioxide tension represents closely the carbon dioxide tension in the arterial blood, it affords a good index of the acidity of the blood. Therefore this index may be much more satisfactory and important than the urinary tests for acidity.

Van Slyke and Cullen (2) have recently adopted the plasma bicarbonate as the ideal measure for the alkaline reserve of the blood; a criterion much better than the measurement of the alveolar carbon dioxide tension for the purpose of determining "acidosis" in the modern broader sense. For this determination they published a new method and designed a suitable apparatus.

TABLE 1

Influence of two extractions

DAYS OF FAST / ANIMAL NUMBER		3 7	4 9	7 16	9 12	11 25
CO_2 in 1 cc. arterial plasma	cc.	0.3642	0.3522	0.3369	0.1723	0.2204
	mgm.	0.7150	0.6911	0.6616	0.3377	0.4328
CO_2 of the same animal preceding day	cc.	0.5170	0.4028	0.4112	0.3960	0.3268
	mgm.	1.015	0.7902	0.8071	0.7778	0.6419

TABLE 2

Influence of second extraction on moribund animals

DAYS OF FAST / ANIMAL NUMBER		5 11	14 28	20 17
CO_2 in 1 cc. of arterial plasma	cc.	0.4486	0.4280	0.4468
	mgm.	0.8812	0.8406	0.8770

TABLE 3

Arterial plasma bicarbonate in short time after death

DAYS OF FAST / ANIMAL NUMBER		12 24	14 26	15 23	18 27
CO_2	cc.	0.5907	0.5837	0.7220	0.8583
	mgm.	1.101	1.1467	1.4182	1.6860

TABLE 4

Arterial plasma bicarbonate during starvation

Days	Animal number	CO_2	CO_2	Animal number	CO_2	CO_2
		cc.	*mgm.*		*cc.*	*mgm.*
Normal	10	0.5495	1.078	18	0.5666	0.113
Days of fast						
1	20	0.4938	0.9690			
2	7	0.5170	1.015			
3	9	0.4028	0.7902			
4	11	0.4096	0.8040			
5	15	0.4088	0.8026			
6	16	0.4112	0.8071			
7	12	0.3960	0.7778			
8						
9	17	0.3830	0.7518			
10	25	0.3268	0.6419			
11	24	0.3423	0.6722			
12						
13	28	0.3469	0.6813			
14						
15						
16	27	0.3723	0.7311	21	0.2720	0.5141

As was shown by several authors, it is doubtless a fact that in the blood during fasting an acidosis develops. I have wished to determine the acidosis precisely by Van Slyke and Cullen's method and found in the blood plasma of 28 rabbits a similar fall of bicarbonate as reported by Benedict (3) on the finding of Levanzin's alveolar air.

This research was begun in January, 1918, at the Institute for Forensic Medicine of the Tokyo Imperial University and concluded in May, 1918. A preliminary report of the work was made at the Forensic Medical Department of the Fifth Japanese Medical Congress in April, 1918. I desire at this time to express my hearty thanks to Prof. Dr. K. Katayama and Prof. Dr. S. Mita for their kind assistance in the direction of my experiments.

MATERIAL AND TECHNIQUE

I collected the arterial blood by means of a cannula tube, allowing it to flow into potassium oxalate solution under a layer of paraffin oil. The oxalate solution was of strength such that the salt amounted to about 0.5 per cent of the blood in the tube. The blood thus drawn

was centrifuged immediately and 1.0 cc. of the plasma was subjected to the determination, according to Van Slyke and Cullen's description. The determination was carried out two or three times on one and the same sample and the average of these was adopted as a result.

RESULTS

Of 28 rabbits used in these experiments, the results of nos. 1 to 6 were discarded because of technical errors. Nos. 8, 13, 14, 19, 21 and 22 died in the course of the fast, consequently their blood plasma bicarbonate could not be determined. The blood of nos. 23 and 26 was taken immediately after their death and subjected to determination. The remainder, i.e., 14 rabbits, were examined before death. The plasma bicarbonate is remarkably influenced by the condition of the animals as the following results will show.

TABLE 5

Loss of body weight

DAYS OF FAST	NUMBER OF ANIMALS						
	9	11	15	16	19	7	8
1	2024	2328		2150	2000		1924
2	1910	2190	2012	2134	1848	*1518*	*1820*
3	*1860*	2150	1940	2126	1764	**1380**	Dead
4	**1790**	*2075*	1872	1934			
5		**2000**	*1812*	1920	Dead		
6			1764	*1904*			
			Dead				
7				**1904**			

Italic figures indicate the first extraction of blood and heavy face figures the second extraction. Animals were not permitted to survive the second bleeding.

1. Influence of various conditions on the arterial plasma bicarbonate

a. Two or more extractions of blood from carotid. As seen in table 1, the plasma bicarbonate is enormously reduced by the second bleeding in animals yet capable of surviving for several days. But in the case of an animal moribund from inanition, the plasma bicarbonate, on the contrary, is more or less increased, as shown in table 2.

b. In short time after death. Shortly after death the arterial plasma bicarbonate definitely increased as compared with the moribund state. This applies equally for the first as for the second bleeding, as is evident from table 3.

2. The amount of plasma bicarbonate in the arterial blood of fasting rabbits

As seen in table 4, the arterial plasma bicarbonate is about 55 volume per cent before the fast, about 50 on the first and second days of the fast, and from the third to the ninth day it falls to a level of about 40 volume per cent; afterwards there occurs a third rather moderate fall, namely, to about 34 volume per cent which remains without any further change until the sixteenth day, when death usually occurs.

3. Loss of body weight during the fast

As presented in tables 5 and 6, the body weight of most experimental animals is seen to be remarkably reduced on the second day of the fast, but the rate of the weight loss is subjected to an extreme fluctuation on account of individual variations and is not necessarily influenced by drawing of blood. The 11 rabbits in table 6, i.e., nos. 12, 17 and 20 to 28 were left in the state of absolute fast until death. They lived for 11 to 20, in the average 14.5 days and lost in the end 27.69 to 52.37, in the average 41.78 per cent of their initial body weight, the same as recorded by several authors. There seems to exist no special relationship between the loss of body weight and the fall in plasma bicarbonate. The more or less conspicuous loss of body weight on the second day of the fast which was often but not always observed, seemed to have some relationship with the first fall in arterial carbon dioxide tension; the second and third falls, however, were accompanied by no precipitate loss of weight.

4. Finding by autopsy

Although the experimental animals lost markedly in weight, the stomachs were invariably found to contain a considerable amount of grayish fecal material. Gross changes were seldom apparent in the organs; coccidosis of liver and intestinal ulcers with bleeding was noted in no. 24, and intra-muscular hemorrhages were found in no. 16. The urine was usually clear and acid to litmus.

Microscopically I found intensive congestion in small arteries and capillaries of every organ (brain, lung, liver, spleen and kidney) and cloudy swelling, vacuolarization and atrophy of various degree in the parenchymatous cells of liver and kidney. None of my preparations show fatty degeneration except in ovary and adrenal. Hemosiderosis of liver cells and pigmentation of spleen were strikingly obvious in animals after 10 days' fast.

TABLE 6

Loss of body weight in more than ten days' fast

DAYS OF THE FAST	NUMBER OF ANIMAL																					
	12		17		20		21		22		23		24		25		26		27		28	
	Weight	Loss	Weight	Loss	Weight	Loss	Weight	Loss	Weight	Loss	Weight	Loss	Weight	Loss	Weight	Loss	Weight	Loss	Weight	Loss	Weight	Loss
1	2290	102	2250	158	*2118*	54	2006		1862	38	1900	72	1878	132	1690	138	1620	100	1860	76	1772	54
2	2188	78	2092	46	2064	140			1824	62	1828	40	1740	38	1552	18	1520	46	1784	68	1718	144
3	2110	46	2046	42	1924				1762	62	1788	48	1702	82	1534	108	1474	46	1716	60	1574	106
4	2064	26	2004	38			*1782*	56	1700	58	1740	16	1620	70	1426	6	1428	18	1656	46	1468	66
5	2038	56	1966	30			*1726*	46	1642	56	1724	74	1550	60	1420	12	1410	70	1610	90	1402	62
6	1982	66	1936	42			*1680*	50	1586	48	1650	20	1490	82	1408	118	1340	40	1520	38	1340	14
7	*1916*	60	1894	34	*1688*	38	*1630*	64	1538	46	1630	60	1408	68	1290	14	1300	40	1482	130	1326	54
8	1856	76	1860	28	*1650*	46	*1566*	60	1482	58	1570	26	1340	84	1276	76	1260	52	1352	46	1272	32
9	*1780*	46	*1832*	58	*1604*	50	*1506*	38	1424	60	1544	52	1256	90	1200	60	1208	48	1306	46	1240	40
10	1734	78	1774		1554	44	1468	44	1364	84	1492	92	1166	112	*1140*	136	1160	52	1260	40	1200	16
11	1656				1510	58	1424	36	1280	86	1400	60	*1054*	74	*1004*		1108	48	1220	30	1084	54
	Dead														Dead							
12					1452	48	1388	14	1194		1340	70	980				1060	68	1190	40	1030	76
													Dead									
13					1404	70	1374	44	Dead		1270	62					992	32	1150	40	*954*	54
14			1560	34	1334	64	1330	54			1208	28					*960*		1110	20	*900*	
																	Dead				Dead	
15			1526	46	1260	22	1276	64			*1180*								1090	30		
											Dead											
16			1480	58	1238		*1212*												*1060*	88		
							Dead															

17			1422	70	Dead												972	86		
18			1352	76													886			
																	Dead			
19			1276	68																
20			*1208*																	
			Dead																	
Actual loss of weight.	634		1042		880		794		668		720		898		686		660		974		872	
Percentage loss.....	27.69		46.30		41.56		39.59		35.88		37.88		47.82		40.59		40.74		52.37		49.20	

Italic figures indicate blood drawing.

CONCLUSIONS

1. On the first and second days of starvation, the plasma bicarbonate in the arterial blood of rabbits showed a drop from the normal value. On the third day of the fast there was a second rather sharp fall, after which there was no change until the ninth day. On the tenth fasting day there occurred a third rather moderate fall, after which no further marked change took place until the end of life. Generally the arterial plasma of rabbits has 55 volume per cent of carbon dioxide. The first fall is about 5 volume per cent, the second about 10 and the third about 6 volume per cent.

2. The amount of carbon dioxide in the arterial plasma is influenced considerably by the condition of the animals. After one extraction of 10 cc. blood from the carotid the acidosis seems to be conspicuously increased, because by the second extraction the amount of carbon dioxide is always less than that of the first extraction on the same fasting day.

In the moribund state a contrary result is obtained, i.e., the amount of carbon dioxide in the arterial plasma does not decrease but increases. This is also the case in the arterial plasma immediately after death. This increase is, I think, not due to bicarbonate, but rather to an accumulation of carbon dioxide caused by the failure of the circulatory as well as the respiratory functions.

3. The rate of the loss of body weight is subjected to wide individual fluctuations which may or not may be influenced by the blood extraction. The animals lived in the state of an absolute fast for 11 to 20, in the average 14.5 days and at the end of life had lost 27.69 to 52.37, in the average 41.78 per cent of their initial weight.

There is no demonstrable relationship between the loss of body weight and the fall in amount of plasma bicarbonate.

4. Microscopically many organs showed cloudy swelling, vacuolarization and atrophy. There was invariably an intensive congestion in every glandular organ, but fatty degeneration was found almost in no case.

BIBLIOGRAPHY

(1) BENEDICT: Carnegie Inst. of Washington Publ., 1907, 438.
(2) VAN SEYKE AND CULLEN: Journ. Biol. Chem., XXX, 289.
(3) BENEDICT: Carnegie Inst. of Washington Publ., 1915, 124.

STUDIES ON THE EXTRACT OF LUNG

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INTRODUCTION

Brieger and Uhlenhuth (1) reported that the hypodermic injection of tissue extract into guinea pigs caused death. More recently Kraus and Volk (2) stated that the intravenous injection of the extract of tuberculous tissue killed guinea pigs. Dold (3), (4) reported further that the extract of normal lung, injected intravenously into rabbits and guinea pigs, caused death and it was also found by him that the toxin involved in this reaction was neutralized by normal fresh serum. Since then various authors (5), (6), (7), (8), (9), (10), (11), (12), (13), (14), (15), (16) have studied the nature and property of this tissue toxin, but their results have been very contradictory. Thus it may be suspected that the tissue toxin in itself is not a simple body. I have investigated the toxin-neutralizing power of a number of substitutes and also studied the influence of the toxin upon the sugar content of the blood.

METHOD OF PREPARING THE LUNG EXTRACT

a. The extract of beef lung. From one to two hours after death, the lung was minced and five parts in weight were mixed to seven volumes of 0.85 per cent sodium chloride solution and allowed to stand two hours in the cold. Then it was passed through lint and the filtrate was centrifuged until the supernatant fluid was free of solid matter. This supernatant fluid was used as the lung extract.

b. The extract of the lungs of rabbits and guinea pigs. Immediately after the death caused by bleeding the lung was taken out and was extracted in the same way as above.

SYMPTOMS OF INTOXICATION IN GUINEA PIGS

All the experiments on the toxic action of the lung extract were carried out on guinea pigs and the injection of the extract was done

into the external jugular vein. When a lethal dose of the extract was thus given intravenously a large majority of the animals were much stimulated, urinated and stooled, had a general clonic convulsion, after which the respiration was labored and in two or three minutes coma developed followed shortly by death. In other cases the symptoms were not so typical; the latent period of the reaction was prolonged and death did not follow for an hour or two hours. When sublethal doses were given, some of the animals were thrown into convulsions, which were followed by labored respiration as above and then after a few minutes to many hours were completely restored; others showed only a transitory dyspnoea. Subcutaneous and intraperitoneal injections produced no acute symptoms.

TOXICITY OF LUNG EXTRACT

The minimal lethal dose of beef lung extract was determined upon guinea pigs by intravenous injection. The extract was always freshly made. In the table (table 1) the notation (+) indicates death within one hour; the notation (−), failure of the dose to kill. Because of variability in the resistance of guinea pigs, each dose was tested on two animals and the minimal dose sufficient to kill both animals was taken as the minimal lethal dose. In the table this has been expressed in cubic centimeters per 100 grams body weight.

As table 1 shows, the toxin varied in potency, the minimal lethal dose on guinea pigs with intravenous injection ranging from 0.02 to 0.15 cc. per 100 grams of the body weight.

ON THE INFLUENCE OF GLUCOSE UPON THE TOXICITY OF THE LUNG EXTRACT

The observation by Dold that a fresh serum neutralized the tissue toxin has been confirmed by many authors. In this connection I undertook to study the influence of 10, 15 and 42 per cent solutions of grape sugar upon the potency of the toxin. In these experiments glucose and extract of beef lung were mixed in vitro in the proportions shown in the following table (table 2) and injected into the external jugular vein of guinea pigs.

TABLE 1

The toxicity of the beef lung extract on guinea pigs

DATE (1918)	BODY WEIGHT	SEX	AMOUNT OF EXTRACT INJECTED PER 100 GRAMS OF BODY WEIGHT	RESULT	TIME FROM INJECTION TO DEATH
	grams		*cc.*		*minutes*
July 9	380	♂	0.06	+	8
	400	♂	0.03	+	6
	380	♀	0.03	+	50
	410	♂	0.02	+	5
	380	♂	0.02	+	12
	390	♂	0.02	−	
	400	♂	0.01	−	
July 11	450	♀	0.02	−	
	370	♀	0.02	−	
	370	♂	0.02	−	
	350	♂	0.03	+	50
	320	♀	0.03	+	6
July 17	440	♀	0.03	−	
	600	♀	0.03	−	
	610	♀	0.03	−	
	640	♀	0.04	−	
	570	♀	0.04	−	
	550	♀	0.05	−	
	610	♀	0.05	−	
	830	♀	0.1	+	18
	500	♀	0.1	+	12
	580	♀	0.1	+	38
July 20	430	♂	0.03	−	
	450	♀	0.03	−	
	470	♀	0.05	+	25
	510	♀	0.05	+	16
	450	♂	0.1	+	17
August 9	400	♂	0.01	−	
	510	♀	0.01	−	
	610	♀	0.02	+	11
	520	♂	0.02	+	15
	490	♂	0.05	+	3
August 10	300	♀	0.05	−	
	400	♂	0.08	−	
	500	♀	0.1	+	3
	570	♀	0.1	+	45

TABLE 1—*Concluded*

DATE (1918)	BODY WEIGHT	SEX	AMOUNT OF EXTRACT INJECTED PER 100 GRAMS OF BODY WEIGHT	RESULT	TIME FROM INJECTION TO DEATH
	grams		*cc.*		*minutes*
August 12	410	♂	0.02	−	
	420	♂	0.05	−	
	420	♂	0.05	+	10
	420	♂	0.1	+	15
	550	♀	0.1	+	45
	380	♂	0.15	+	55
	440	♀	0.15	+	5
	490	♀	0.2	+	9
September 2	400	♀	0.1	+	5
	500	♀	0.05	+	13
	290	♀	0.05	+	50
	520	♀	0.02	+	5
	500	♀	0.02	+	5
	500	♂	0.01	−	
	570	♀	0.01	−	
September 11	400	♀	0.02	−	
	500	♂	0.02	+	9
	490	♀	0.05	+	6
	460	♂	0.05	+	55
	450	♀	0.08	+	5
	520	♀	0.1	+	4
	360	♀	0.1	+	20
September 14	510	♀	0.05	−	
	440	♂	0.1	−	
	360	♀	0.1	−	
	360	♂	0.15	+	3
	420	♀	0.15	+	7
September 17	400	♂	0.05	−	
	380	♂	0.1	+	55
	420	♂	0.1	+	40
	450	♂	0.15	+	34
	510	♀	0.15	+	19
	390	♀	0.2	+	4

TABLE 2

The influence of glucose solution upon the toxicity of the beef lung extract on guinea pigs

DATE (1918)	BODY WEIGHT	SEX	AMOUNT OF GLUCOSE MIXED	AMOUNT OF NaCl SOLUTION (0.85 PER CENT) MIXED	AMOUNT OF LUNG EXTRACT PER 100 GRAMS OF BODY WEIGHT	RESULT	TIME FROM INJECTION TO DEATH
	grams		*cc.*	*cc.*	*cc.*		*minutes*
July 11	350	♂			0.03	+	50
	320	♀			0.03	+	6
			10 per cent solution				
	400	♂	0.5		0.03	+	8
	400	♀	1.0		0.03	−	
	380	♀	1.5		0.03	−	
July 11	310	♂	1.5		0.45	−	
	360	♀	1.5		0.06	+	26
	370	♂	1.5		0.09	+	20
	390	♂		1.0	0.03	+	50
	520	♀			0.02	+	5
	500	♀			0.02	+	5
	430	♂	0.5		0.02	+	12
September 2	440	♀	1.0		0.02	−	
	400	♀	1.5		0.02	−	
	390	♂	1.5		0.03	−	
	410	♀	1.5		0.05	+	7
			15 per cent solution				
	390	♂	0.5		0.03	−	
	320	♂	1.0		0.03	−	
	350	♂	1.5		0.03	−	
	350	♀	1.5		0.045	−	
July 11	350	♂	1.5		0.06	−	
	370	♀	1.5		0.09	+	20
	360	♂	1.5		0.175	+	15
	350	♂			0.03	+	50
	320	♀			0.03	+	6
	470			1.0	0.03	+	32
	520	♀			0.02	+	5
	500	♀			0.02	+	5
	470	♂		1.0	0.02	+	55
September 2	340	♂	0.5		0.02	−	
	380	♂	1.5		0.06	−	
	370	♀	1.5		0.1	−	
	490	♀	1.5		0.15	+	14

TABLE 2—*Continued*

DATE (1918)	BODY-WEIGHT	SEX	AMOUNT OF GLUCOSE MIXED	AMOUNT OF NaCl SOLUTION (0.85 PER CENT) MIXED	AMOUNT OF LUNG EXTRACT PER 100 GRAMS OF BODY WEIGHT	RESULT	TIME FROM INJECTION TO DEATH
	grams		*cc.*	*cc.*	*cc.*		*minutes*
			42 per cent solution				
August 17	830	♀			0.1	+	18
	500	♀			0.1	+	13
	580	♀			0.1	+	38
	560	♀	0.5		0.1	+	43
	500	♀		0.5	0.1	+	15
	560	♀	1.0		0.1	+	19
	680	♀	1.5		0.1	+	60
	370	♀		1.5	0.1	+	20
	590	♀	2.0		0.1	−	
	570	♀	2.5		0.1	−	
	580	♀		2.5	0.1	+	40
	520	♂	3.0		0.1	−	
	500	♂		3.0	0.1	+	13
September 2	520	♀			0.02	+	5
	500	♀			0.02	+	5
	460	♀	0.5		0.02	−	
	300	♂	1.0		0.02	−	
	380	♂	0.5		0.05	+	10
	550	♂	1.5		0.02	−	
	330	♀	1.5		0.08	−	
	260	♀	1.5		0.1	−	
	400	♂	1.5		0.1	−	
	420	♀	1.5		0.12	+	7
	380	♂	1.5		0.12	−	
September 11	490	♀			0.05	+	6
	460	♂			0.05	+	55
	550	♂	0.5		0.05	+	8
	640	♂	1.0		0.05	+	18
	530	♂	1.0		0.05	−	
	490	♀	1.5		0.05	−	
	600	♀	1.5		0.1	−	
	540	♀	1.5		0.1	+	17
	500	♂	1.5		0.15	+	28

TABLE 2—*Concluded*

DATE (1918)	BODY WEIGHT	SEX	AMOUNT OF GLUCOSE MIXED	AMOUNT OF NaCl SOLUTION (0.85 PER CENT) MIXED	AMOUNT OF LUNG EXTRACT PER 100 GRAMS OF BODY WEIGHT	RESULT	TIME FROM INJECTION TO DEATH
	grams		*cc.*	*cc.*	*cc.*		*minutes*
September 14.	360	♂			0.15	+	11
	420	♀			0.15	+	7
	430	♀	0.5		0.15	+	10
	540	♂	1.0		0.15	−	
	420	♀	1.0		0.15	−	
	500	♂	1.5		0.15	−	
	500	♂	1.5		0.15	−	
	520	♀	1.5		0.25	−	
	370	♂	1.5		0.3	+	24
	380	♀	1.5		0.3	−	
September 17.	420	♂			0.1	+	40
	380	♂			0.1	+	55
	410	♀	0.5		0.1	+	23
	390	♂	1.0		0.1	+	16
	400	♂	1.5		0.1	−	
	410	♂	1.5		0.2	−	
	400	♂	1.5		0.2	−	
	420	♀	1.5		0.3	+	14

The results obtained in the above experiments may be summarized as following:

1. One cubic centimeter of 10 per cent solution of glucose is effective to protect guinea pigs against the minimal lethal dose of the beef lung extract and 1.5 cc. of the same solution protects against 1.5 times the lethal dose.

2. Five-tenths cubic centimeter of 15 per cent solution is also effective against the minimal lethal dose and 1.5 cc. against 2 to 3 times the lethal dose.

3. Five-tenths to one cubic centimeter of 42 per cent solution is effective against the minimal lethal dose and 1.5 cc. against 1.5 to 5.0 times the minimal lethal dose.

Therefore it is certain that glucose is effective in protecting guinea pigs against the beef lung extract, but the nature of such action is still obscure.

TABLE 3

The influence of adrenalin upon the toxicity of the beef lung extract for guinea pigs

DATE (1918)	BODY WEIGHT	SEX	AMOUNT OF ADRENALIN INJECTED	AMOUNT OF EXTRACT INJECTED PER 100 GRAMS OF BODY WEIGHT	RESULT	TIME FROM INJECTION TO DEATH
	grams		*cc.*	*cc.*		*minutes*
August 12	420	♂		0.05	−	
	420	♂		0.05	+	10
	550	♀		0.1	+	45
	420	♂		0.1	+	15
	390	♂	0.3	0.1	−	
	570	♂	0.2	0.1	−	
	380	♂		0.15	+	55
	440	♀		0.15	+	5
	450	♂	0.2	0.15	−	
	560	♂	0.2	0.15	−	
	490	♀		0.2	+	9
	450	♀	0.3	0.2	+	30
	570	♀	0.3	0.2	+	24
	360	♀	0.2	0.2	+	20
	470	♂	0.2	0.2	+	16
September 2	520	♀		0.02	+	5
	500	♀		0.02	+	5
	300	♀	0.2	0.02	−	
	450	♀	0.2	0.02	−	
	350	♂	0.2	0.03	−	
	390	♂	0.2	0.04	+	8
	330	♂	0.2	0.04	−	
	340	♀	0.2	0.05	+	25
	390	♂	0.2	0.05	−	
	340	♀	0.2	0.06	+	20
	390	♂	0.2	0.06	−	
September 11	490	♀		0.05	+	6
	460	♂		0.05	+	55
	540	♀	0.2	0.05	−	
	400	♂	0.2	0.1	+	15
	390	♂	0.2	0.1	+	27
	580	♀	0.2	0.1	−	
	350	♀	0.2	0.15	+	8
September 14	360	♂		0.15	+	3
	420	♀		0.15	+	7
	520	♀	0.2	0.15	+	5
	480	♂	0.2	0.15	−	

TABLE 3—*Concluded*

DATE (1918)	BODY WEIGHT	SEX	AMOUNT OF ADRENALIN INJECTED	AMOUNT OF EXTRACT INJECTED PER 100 GRAMS OF BODY WEIGHT	RESULT	TIME FROM INJECTION TO DEATH
	grams		*cc.*	*cc.*		*minutes*
September 17	410	♀	0.3	0.15	−	
	370	♀	0.2	0.25	+	39
	330	♀	0.2	0.25	+	17
	470	♂	0.3	0.3	+	4
	390	♀	0.2	0.3	−	
September 17	420	♂		0.1	+	40
	380	♂		0.1	+	55
	390	♀	0.2	0.1	−	
	420	♂	0.2	0.1	+	40
	440	♀	0.2	0.15	−	
	430	♂	0.2	0.15	+	21
	440	♂	0.2	0.15	+	12
	430	♀	0.2	0.2	+	28

ON THE INFLUENCE OF ADRENALIN UPON THE TOXIC EFFECT OF THE EXTRACT

There are many clinical and experimental publications on the toxin-neutralizing power of adrenalin, but all these reports do not harmonize and the nature of the neutralizing action is yet unknown. The following experiments have been made to determine the biological influence of adrenalin on the extract of the beef lung.

After the hypodermic injection of 0.2 to 0.3 cc. of adrenalin (adrenalin chloride, 1: 1000, Sankyo and Co.) the extract of beef lung was immediately injected into the external jugular vein of guinea pigs.

From table 3 it is to be seen that the hypodermic injection of adrenalin (0.2 to 0.3) is effective to protect guinea pigs against 1 to 1.5 times the minimal lethal dose of the beef lung extract.

ON THE INFLUENCE OF THE INTRAVENOUS INJECTION OF THE LUNG EXTRACT UPON THE SUGAR CONTENT OF THE BLOOD

After studying the neutralizing and protective effects of glucose and adrenalin against the lung extract, the influence of the toxin upon the blood sugar was taken up for investigation. It is well known that

hyperglycemia is caused by injection of various toxins. The lung extract of guinea pigs was injected intravenously into rabbits and the blood sugar was determined by Bang's micromethod. In this series of experiments, rabbits were mostly used and special precautions were taken to avoid other influencing factors.

TABLE 4

The influence of the lung extract of guinea pigs on the sugar content of the blood of rabbits

BODY WEIGHT	SEX	AMOUNT OF EXTRACT INJECTED	BLOOD SUGAR CONTENT (PER CENT)									
			Before injection	Hours after injection								
				½	1	1½	2	2½	3	4	5	6
grams		*cc.*										
2740	♂	1.5	0.132	0.127	0.137	0.153	0.155	0.150	0.182	0.150	0.149	
1790	♂	1.5	0.120	0.113	0.114	0.104	0.103	0.099	0.116	0.122	0.119	
1680	♀	2.0	0.095	0.084	0.081	0.101	0.111	0.110	0.118	0.114	0.101	
2370	♂	2.0	0.104	0.105	0.111	0.112	0.108	0.118	0.131	0.103	0.102	0.104
2560	♀	1.5	0.118	0.100	0.110	0.115	0.128	0.119	0.135	0.121	0.119	

This table shows that the injection of the lung extract causes a slight increase in the blood sugar.

SUMMARY

1. Intravenous injection of beef lung extract in guinea pigs invariably causes dyspnoea and, in a majority of cases, convulsions as well.

2. The minimal lethal dose of the beef lung extract for guinea pigs on intravenous inoculation varied from 0.02 to 0.15 cc. per 100 grams of body weight.

3. Glucose destroys the toxicity of lung extracts. Thus 1.0 cc. of 10 per cent glucose mixed with the minimal lethal dose of lung extract renders the latter inert. Additional figures are given in the text.

4. Hypodermic injection of adrenalin immediately before an intravenous lethal dose of lung extract was protective in effect.

5. Intravenous injection of lung extract causes a slight increase in the blood sugar of rabbits.

I take this opportunity to express my gratitude to Prof. Dr. S. Mita for his suggestions, advice and criticism in carrying out this investigation.

BIBLIOGRAPHY

(1) Brieger und Uhlenhuth: Deutsch. med. Wochenschr., 1898, no. 10.
(2) Kraus und Volk: Wiener klin. Wochenschr., 1910, no. 8.
(3) Dold: Zeitschr. f. Immunität., 1911, x.
(4) Dold: Deutsch. med. Wochenschr., 1911, no. 36.
(5) Aronson: Berl. klin. Wochenschr., 1913, no. 6.
(6) Aronson und Izar: Münch. med. Wochenschr., 1912, no. 20.
(7) Dold und Ogata: Zeitschr. f. Immunität., 1912, xiv.
(8) Dold und Kodama: Zeitschr. f. Immunität., 1913, xviii.
(9) Gutmann: Zeitschr. f. Immunität., 1913, xix.
(10) Izar und Patane: Zeitschr. f. Immunität., 1912, xiv.
(11) Ichikawa: Zeitschr. f. Immunität., 1913, xviii.
(12) Goto: Organ d. med. Gesellsch. zu Kyoto, xiii, 1.
(13) Goto: Organ d. med. Gesellsch. zu Kyoto, xiii, 3.
(14) Yoshimura: Nippon Biseibutsugakkai Zashi, ii.
(15) Ishikawa: Ber. d. med. Gesellsch. zu Tokyo.
(16) Obata: Mitteil. d. med. Gesellsch. zu Tokyo, 1916, xxx, no. 22.

VITAL STAINING AND ACIDOSIS

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INTRODUCTION

The vital staining method by injection of lithium carmine solution into the ear vein of living animals has recently been applied to investigations in the field of medicine and biology. But the chemical reaction between the stain and the living cells is as yet obscure. Many authors assume some specific granula in the protoplasm of the living cells which serve to take up the stain. Kiyono (1), (2) assumes their increase in the supravital state from the fact that carmine granula taken up by surviving cells are always more in number and finer in size. Though nearly all living cells take carmine stains, the epithelium of the bile-duct is never found to be stained because, presumably, of its alkaline reaction, while cells of brain, plexus chorioideus ventriculi quarti and the intestinal mucous membrane take the stains in a trace and in a state of the finest granula, again presumably on account of their rather alkaline reaction. We know also that the carmine stains are quite toxic to the living cells and because of this an ideal staining requires one administration a day in a not very toxic dose, at least for several days. From these data I would like to conceive that the carmine granula are not due to staining of the specific intracellular granula, but are due rather to a physical and physico-chemical phenomenon, which takes place as a consequence of acidosis in the organism caused by the carmine injection.

This research was begun for the purpose of testing this hypothesis in October, 1917, and concluded in April, 1918, at the Institute for Forensic Medicine of the Tokyo Imperial University. It was presented before the Pathological Section of the Fifth Japanese Medical Congress held in April, 1918, in Tokyo. In connection with this report I acknowledge my pleasant obligation to express gratitude to Prof. Dr. K. Katayama and Prof. Dr. S. Mita for their kind direction and helpful advice.

EXPERIMENTAL

First I demonstrated the precipitation of carmine granula from the lithium carmine in a very dilute solution of hydrochloric acid in vitro, secondly the acidosis in the blood of guinea pigs in which the stains were intraperitonally administered, and thirdly I realized a more conspicuous staining in animals in which acidosis was previously called forth by various manipulations.

The method carried out and the results obtained were as follows:

1. Precipitation of carmine granula from the lithium carmine solution made acid by the addition of a very small amount of hydrochloric acid. I dissolved 5 grams of the purest carmine (Grübler) in a saturated solution of lithium carbonicum. The reaction of the solution obtained was strongly alkaline, that is, it blued litmus paper sharply. There was never found microscopically any visible granula in the solution after filtration through gauze. I put it into a test tube, diluted it with

TABLE 1

Plasma bicarbonate of normal guinea pigs

	GUINEA PIG			AVERAGE
	6	8	9	
Body weight (grams)	510	510	544	
CO_2 (cc. in 1 cc. plasma)	0.3806	0.4373	0.4082	0.4087

aqua distillata or blood plasma of normal rabbits and microscopically found as before no visible carmine granula in it. But as soon as I added some drops of 0.1 per cent solution of hydrochloric acid the color of the carmine red solution turned a little pale, and in a sample of it many red granula became microscopically visible. After 24 hours the solution was separated in two different parts: the greater upper one consisting of clear colorless watery fluid and the smaller lower of a red sediment. I observed the similar precipitation also by addition of various electrolytes, e.g., ammonium sulphate, calcium chloride, silver chloride, etc., and also of various acids, but it did not happen when sodium bicarbonate, sodium chloride, etc., were added; in the case of Ringer's solution it became purplish without, however, any precipitation. The lithium carmine solution was dialysable through the thimble (Carl Schleicher and Schüll) extraordinarily slowly.

It is evident from these in-vitro experiments that carmine is capable of dissolving in alkaline lithium carbonicum solution in a state of col-

loidal solution, but is liable to precipitation in a slightly acidified solution or a slight excess of hydrogen ion concentration on the one hand and also in the presence of electrolytes of an easily dissociable sort on the other hand.

2. Acidosis in the blood plasma of guinea pigs stained vitally with lithium carmine. I injected intraperitoneally 1 or 2 cc. of 5 per cent lithium carmine solution, after sterilization and filtration through gauze, into guinea pigs once a day for more than ten days. I collected

TABLE 2

Body weight loss of guinea pigs in the course of vital staining with lithium carmine solution

DAYS OF EXPERIMENT	GUINEA PIG			AMOUNT OF CARMINE INJECTED
	3	4	5	
	Body weight	Body weight	Body weight	
	grams	*grams*	*grams*	*cc.*
1	590	435	550	1.0
2	570	435	560	1.0
3	590	435	590	1.0
4	610	400	615	1.0
5				
6	620	428	600	1.0
7	636	416*	620	2.0
8	600	400	560	2.0
9	530	410	530	2.0
10	500	380	500	2.0
11	480	360	476	2.0
12				
13	420†			
14			390†	

* Blood drawn.

† Blood drawn and killed.

the arterial blood from the carotid by help of a cannula into a paraffined centrifuge tube containing a layer of paraffin oil and provided with potassium oxalate to make about 0.5 per cent the weight of the blood drawn, under precautions to prevent its free contact with air and to avoid any agitation. The blood thus drawn was centrifuged immediately and 1 cc. of the plasma was subjected to Van Slyke and Cullen's determination of bicarbonate. The determination was carried out two or three times on the one and the same sample and their average figure

was adopted as a result. As two or more bleedings reduce the amount of plasma bicarbonate, as I have previously shown in fasting rabbits (4), the first blood sample only was used to the present experiments.

a. Plasma bicarbonate of normal guinea pigs. As seen from the table 1, in guinea pigs the plasma bicarbonate was determined at 40.87 volume per cent, i.e., much less than about 60, the value in man, rabbits and many other animals. It was, I think, because of their far more vivacious motion than that of rabbits, etc., before being attached to the fastening apparatus.

b. Body weight loss of guinea pigs in the course of vital staining with lithium carmine. As shown in table 2, the intraperitoneal administration of 1 cc. lithium carmine to three guinea pigs of 435 to 590 grams, i.e., of 0.19 cc. per 100 grams seems not to affect their health, but that of 2 cc. to the animals of 416 to 636 grams, i.e., of 0.358 cc. per 100 grams, markedly reduces the body weight.

TABLE 3

Plasma bicarbonate of guinea pigs after vital staining

	GUINEA PIG		
	3	4	5
Day of blood drawing	13th	7th	14th
CO_2 (cubic centimeter)	0.2354	0.3109	0.1573
CO_2 (milligram in 1 cc. plasma)	0.4625	0.6066	0.3082

Therefore it may be said that 5 per cent lithium carmine has a toxic action when more than 0.35 cc. per 100 grams is intraperitoneally administered to guinea pigs.

c. Plasma bicarbonate of guinea pigs after vital staining with 5 per cent lithium carmine solution. These results are presented in table 3. Taking 40.87 per cent as the normal value for the arterial plasma bicarbonate, it will be seen that the treatment with lithium carmine solution has a very decided effect. No. 4, on the seventh day of the experiment, with an insignificant loss in body weight, gave 31 per cent plasma bicarbonate. No. 3, on the thirteenth day of the experiment, two days after the injections had been stopped, showed a loss in body weight of 34 per cent and a plasma bicarbonate of only 23.54 per cent. In no 5 the acidosis was so far advanced on the fourteenth day of the experiment, i.e., three days after the final carmine injection, that the plasma bicarbonate amounted to only 15.73 per cent. At this time the animal had lost 37 per cent of its body weight.

TABLE 4

Preinjection of alkali or acid before carmine injection

	DATE	GROUP I			GROUP II			GROUP III	
		Rabbit 1	Rabbit 6	Rabbit 7	Rabbit 3	Rabbit 4	Rabbit 8	Rabbit 2	Rabbit 5
Body weight	1	1986	2040	1528	1926	1898	1830	1558	2418
	2	2144	2170		1868	1896		1530	2660
	3	2036	2100		1900	1756		1490	2466
	4	2012	2156		1818	1800		1532	2480
	5					1808			2588
Preinjection	1	$\frac{N}{10}$ HCl, 20 cc.	$\frac{N}{10}$ HCl, 10 cc.	$\frac{N}{30}$ HCl, 45 cc.	$\frac{N}{30}$ $NaHCO_3$, 10 cc.	$\frac{N}{10}$ $NaHCO_3$, 19 cc.	$\frac{N}{30}$ $NaHCO_3$, 54 cc.	Without preinjection	Without preinjection
	2	10 cc.	20 cc.		10 cc.	8 cc.			
	4		$\frac{N}{30}$ HCl, 40 cc.			$\frac{N}{30}$ $NaHCO_3$, 40 cc.			
Carmine Injection	1			8.0 cc.			9.0 cc.		
	3	5.0 cc.	5.0 cc.		5.0 cc.	5.0 cc.		5.0	5.0 cc.
	4		8.0 cc.			5.0 cc.			6.0 cc.
	Time after preinjection		30′	50′		30′	40′		

Fats.....	1			About 1.5 hours after staining killed			About 1 hour after staining killed		
	4	Killed	Immediately after staining dead with cramps		Killed			Killed	
	5					Killed			Killed
Urine in bladder	Reaction	Slightly alkaline	Amphoteric	Slightly acid	Acid	Slightly acid	Alkaline	Amphoteric	Slightly alkaline
	Color, etc.	Red turbid	Slightly turbid	Carmine red	Red clear	Slightly turbid carmine red	Carmine red turbid	Reddish	Reddish intensively turbid

Since the intraperitoneal administration of 0.19 cc. of 5 per cent lithium carmine per 100 grams body weight for six days gave incomplete vital staining, the dose was increased to 0.358 cc. per 100 grams, and continued for five days more. This yielded a satisfactory stain but left the animal moribund in extreme acidosis and loss of body weight.

3. Vital staining of animals previously made acidotic. In order to call forth acidosis, I injected a slightly acid or a slightly alkaline solution into the ear vein or into the peritoneal cavity of a rabbit or a mouse. It was possible then to vitally stain the animal to a very high degree with a single injection of the carmine solution.

a. Vital staining of rabbits after preinjection of alkali or acid. To this series of experiments, 8 rabbits were used, classifying them into 3 groups; to the first group a tenth or thirtieth normal solution of hydrochloric acid was administered into the ear veins, to the second a tenth or thirtieth normal solution of sodium bicarbonate, and the third remained without any preinjection.

As the blood was very liable to coagulate in the presence of acid, I could not easily inject a sufficient amount of the tenth normal solution. A concentration of a thirtieth normal solution was relatively easily administered. It was the same in the case of alkali. After these preinjections 5 per cent lithium carmine was administered intravenously in intervals of from 30 minutes to 24 hours. The animals thus stained vitally were killed by means of bleeding from carotid at various times after the final injection. At autopsy nearly all the glandular organs were carmine red, but the adrenal was almost always yellowish. In bladder the urine was very interesting: In the urine of No. 7, which was killed about 2 hours after the preinjection of 45 cc. $\frac{N}{10}$ HCl and an hour after the intravenous administration of 8 cc. lithium carmine, carmine granula and bladder epithelia with carmine red nuclei were found abundantly under the microscope. The urine was slightly turbid, slightly acid and of carmine red color. The urine of no. 8, which was killed also about 2 hours after the preinjection of 54 cc. $\frac{N}{30}$ $NaHCO_3$ and an hour after the intravenous administration of 9 cc. lithium carmine, was also carmine red, turbid and of alkaline reaction, and contained no carmine granula nor cells with stained nuclei. The urine of no. 4 which was killed about 24 hours after the final preinjection of alkali and carmine administration, was carmine red, turbid and slightly

acid. The urine of no. 6, which died with cramps immediately after the carmine injection to the amount of 8 cc. and 30 minutes after the third preinjection of a thirtieth normal solution of hydrochloric acid amounting to 40 cc., was slightly turbid, slightly red and of amphoteric reaction. The urines of those which were killed about 40 hours after the second preinjection of alkali or acid and about 20 hours after the carmine injection, gave, in general, a reaction the opposite of that of the preinjected solution. The urines of those which were not preinjected with acid or alkali before the carmine staining, remained slightly alkaline or amphoteric. Generally speaking, in a short time after preinjection of acid or alkali, the urine is of nearly normal reaction, but 24 to 40 hours after the preinjection its reaction becomes quite the opposite of the preinjected solution in spite of the alkaline reaction of the carmine which was always administered afterwards.

TABLE 5

Distribution of carmine in body organs of the preinjected rabbits after vital staining

	RABBIT							
	1	6	7	3	4	8	2	5
Distribution of carmine in:								
Liver	±	±	−	±	±	±	±	±
Kidney	−	−	+!	−	−	+!	−	−
Spleen	−	−	−	−	−	−	−	−
Brain	−			−			−	
Heart	−			−			−	

This fact is very interesting in connection with explanation of carmine-acidosis, because the repeated injections of alkaline carmine solution may of itself cause an acidosis in the experimental animals.

At autopsy there were no special changes aside from the carmine staining of the internal organs. But on histological examination of the tissues I found nearly always cloudy swelling, coagulation, vascularization and fatty degeneration of different degree in the liver, kidney, spleen, and some other organs, without any conspicuous difference under the three groups.

With regard to the distribution of carmine granula in various organs also, as shown in table 5, I cannot find any special difference under the three groups. In the kidney of no. 6 which died immediately after the carmine injection, no carmine granula were found, but in that of no. 7 and 8 which were killed in 1 or 1.5 hours after the vital staining, the

TABLE 6

Vital staining of mice after preinjection of acid or alkali

MOUSE NUMBER	PREINJECTION		INTERVAL	STAIN	TIME OF DECAPITATION AFTER STAINING	DISTRIBUTION OF CARMINE				
						Smear			Paraffin	
	Kind	Amount				Blood	Mesenterium	Liver	Liver	Kidney
		cc.		*cc.*						
3	$\frac{N}{30}$ HCl	p. 0.5	30′	v. 0.1	3 hrs.	−		−		
4	$\frac{N}{30}$ HCl	p. 0.5	30′	v. 0.1	3 hrs.	Trace		Trace		
6	$\frac{N}{30}$ HCl	p. 0.5	30′	v. 0.2	1.5 hrs.	−		Trace		
7	$\frac{N}{30}$ HCl	p. 0.5	30′	v. 0.2	1.5 hrs.	−		−		
19	$\frac{N}{30}$ HCl	p. 0.5	7 days	p. 0.5	2 hrs.	−	Trace	−		
32	$\frac{N}{10}$ HCl	p. 0.5	1.5 hrs.	p. 0.5	2 hrs.	−	±	−	Trace	−
33	$\frac{N}{10}$ HCl	p. 0.5	1.5 hrs.	p. 0.5	2 hrs.	−	±	−	±	±
43	$\frac{N}{10}$ HCl	p. 0.5	20′	v. 0.1	20′	−	−	−	−	−
44	$\frac{N}{10}$ HCl	p 0.5	20′	v. 0.1	20′	−	−	−	−	−
45	$\frac{N}{10}$ HCl	p. 0.5	20′	v. 0.1	20′	−	−	−	−	−
1	$\frac{N}{30}$ $NaHCO_3$	p. 0.5	30′	v. 0.1	3 hrs.	−		−		
2	$\frac{N}{30}$ $NaHCO_3$	p. 0.5	30′	v. 0.1	3 hrs.	−		−		
8	$\frac{N}{30}$ $NaHCO_3$	p. 0.5	30′	v. 0.1	3 hrs.	−		−		
9	$\frac{N}{30}$ $NaHCO_3$	p. 0.5	30′	v. 0.1	3 hrs.	−		−		
26	$\frac{N}{10}$ $NaHCO_3$	p. 0.5	30′	p. 0.3	1.5 hrs.	−	Trace	−		
27	$\frac{N}{10}$ $NaHCO_3$	p. 0.3	30′	p. 0.3	1.5 hrs.	−		−		
29	$\frac{N}{10}$ $NaHCO_3$	p. 0.5	1.5 hrs.	p. 0.5	2 hrs.	+	+	+	++	+
30	$\frac{N}{10}$ $NaHCO_3$	p. 0.5	1.5 hrs.	p. 0.5	2 hrs.	+	+	+	++	+
31	$\frac{N}{10}$ $NaHCO_3$	p. 0.5	1.5 hrs.	p. 0.5	2 hrs.	+	+	+	++	+
5	Without			v 0.5	Dead on the spot	++		++		
28	Without			p 0.3	1.5 hrs.	−	Trace	−		
37	Without			v 0.2	2 hrs.	−	−	−		

N. B. The abbreviation v. indicates intravenous injection and p. intraperitoneal one.

staining was conspicuous; abundant in bases of epithelia of the tubili contorti of no. 8 and in traces in lumina of urine-tubules of no. 7.

b. Vital staining of mice after preinjection of acid or alkali. Since rabbits require a large amount of carmine I resorted to the use of mice in order to spare the stains. Their weight was 10 to 15 grams. The preinjections were carried out to the amount of 0.5 cc. for the most part into the peritoneal cavity, and the stain was generally injected into the tail vein to the amount of 0.1 or 0.2 cc.; the intravenous administration of 0.3 cc. of the stain often killed the animals at once, but the intraperitoneal injection of 0.5 cc. was not so harmful.

In from 20 minutes to 3 hours after the vital staining the mice were killed by decapitation, and immediately smear preparations on coverglasses were made from blood and liver on the one hand, and on the other hand pieces of mesentery were stretched out between two coverglasses. These preparations were stained with hematoxylin only or combining eosin thereto, after their fixation in ether and alcohol. Also specimens of liver and kidney of most of the mice were imbedded in paraffin, cut and stained with hematoxylin and hematoxylin and eosin.

Microscopically I found no special difference between the two series, i.e., the animals treated with alkali and acid. Cloudy swelling, coagulation, pycnosis of the nucleus, karyorhexis or karyolysis were found in nearly all preparations. But as shown in table 6, the animals treated with alkali (nos. 29, 30, 31). took the stains conspicuously as compared with those treated throughout in the same manner except for the preinjection of acid (nos. 32, 33), in which only a trace of the stain was found. In the other animals even the carmine distribution did not differ. Those which were vitally stained by intravenous injection of less than 2 cc. carmine within 30 minutes of the preinjection nearly always gave negative results. Those which were vitally stained by intraperitoneal injection of 0.5 cc. carmine an hour and a half after the preinjection, on the other hand, gave positive results.

DISCUSSION

From a perusal of the above facts it may be readily seen that vital staining with lithium carmine produces an acidosis, and that if acidosis be established as a preliminary to the injection of lithium carmine, the staining is much more conspicuous than in animals not so treated.

The relationship between acidosis and vital staining is difficult to explain. The current conception of cloudy swelling (5) is helpful. In vitally stained tissue the association of the carmine granula and cell edema (cloudy swelling) is generally recognized. If the edema is

developed by a rise of intracellular osmotic pressure due to an abnormal splitting of the cellular proteins in the absence of an adequate oxygen supply the carmine solution may be carried in with the production of edema. It is conceivable then that the carmine granula are precipitated in the acid medium, thus completing the histological picture. This will explain the fact that carmine granula are found only in cells with easily permeable walls and which are exposed to a slow blood stream, e.g., the reticulo-endothelial cells or histiocytes, while these granula are never found in those cells which are constantly exposed to alkaline body fluids or are bathed in a rapid blood stream where water and carmine diffuse in hardly more rapidly than the crystalloids diffuse out, e.g., the cells of the brain, bile ducts and greater blood vessels.

We may account, similarly, for the ease in staining surviving cells as opposed to living cells. If coagulation developed as the result of increased hydrogen-ion concentration before the stain could enter, there would be no deposition of carmine granula. Further, even after the cell walls lose their semipermeable character, the nuclei, possessing a relatively high hydrogen-ion concentration and still permeable to the dye, will take the stain, which has already entered the cell, and make the nuclear figures distinct.

SUMMARY

Vital staining can not be accomplished by a single intravenous or intraperitoneal injection of lithium carmine in subtoxic doses in a healthy animal. If, however, an acidosis be first established, a single injection of the dye will give a satisfactory stain.

In animals which have been vitally stained, an actual decrease in plasma bicarbonate occurs.

Hence the conclusion is drawn that it is incorrect to predicate the existence of specific stain-taking substances or granula in the cells. Rather, vital staining with lithium carmine is due to the development of an acidosis which so alters the function of the body cells that the dye diffusing in is deposited in granula. This deposition corresponds to the precipitation from colloidal solution of the dye when the normally alkaline solution is made acid in vitro.

BIBLIOGRAPHY

(1) KIYONO: Die vitale Karminspeicherung, 1914, 7.
(2) KIYONO AND KATSUNUMA: Kyoto Igaku Zassi, xv, 6.
(3) VAN SLYKE AND CULLEN: Journ. Biol. Chem., xxx, 289.
(4) ASADA: This Journal, 1919, I, 1.
(5) WELLS: Chemical pathology, 1918, 394.

STUDIES IN SECONDARY TRAUMATIC SHOCK

IV. The Blood Volume Changes and the Effect of Gum Acacia on their Development

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In our earlier shock studies the hemodynamic findings all pointed directly to a decreased effective blood volume as the only constant factor tending toward failure of the circulation. Attention has also been called by numerous other observers to the significance of the reduction in both the effective and absolute volumes in this condition (1).

The present experiments were designed to determine the rôle that the absolute volume of the blood plays in the reduction of the effective volume in shock, and to evaluate the possible modes by which such a reduction might be brought about. Three possibilities suggest themselves as to ways the blood volume may be reduced: 1, hemorrhage, external or into tissues; 2, concentration of the blood, i.e., by filtration of the plasma; and 3, stasis in a portion of the vascular bed.

Four forms of experimental shock were studied, namely, those produced by injections of massive doses of adrenalin; by clamping the vena cava above the liver for a period of three hours so that the general arterial pressure was 30 to 40 mm. of mercury; by clamping the abdominal aorta above the coeliac axis for a period of three hours so that the distal pressure was 30 mm. of mercury and by exposure and manipulation of the intestines. Dogs were used in all experiments.

External hemorrhage was absolutely excluded except for the small amounts of blood necessary for the samples. The blood pressure was determined by a small bore manometer connected directly to the artery. It was seldom necessary to "wash out" during the course of an experiment, so that the loss of very few cubic centimeters of blood was involved.

Samples were taken directly from a large artery through a clean dry cannula. The red blood cells were counted as an index to the

relative plasma loss and the results obtained, therefore, indicate the minimum loss of blood through filtration of plasma as any local retention of the erythrocytes would decrease the count. In the later experiments the hemoglobin changes were determined by means of the Dubosque colorimeter, the blood being diluted 1/100 with 0.03 per cent HCl, and the normal hemoglobin dilution being considered as 100 per cent.

In case blood is withdrawn from circulation either by hemorrhage into tissue or stasis in a portion of the capillary bed, such a reduction could be appreciated only by a direct determination of the blood volume. Accordingly such determinations were made by the method of Meek and Gasser (2). Determinations may be made by this method to an accuracy within 5 per cent. Four cubic centimeters per kilo of 20 per cent acacia were injected intravenously, and ten minutes after the injection a sample of blood was taken for the distillations. All determinations were done in duplicate. If stasis occurs it might be of a degree varying from a local retardation of the stream to an absolute removal from active circulation. As ten minutes were allowed between injecting and sampling, the acacia would be diluted by all the blood that passes through the general circulation in a period of ten minutes.

In this series the final shock determinations were made after the mean arterial pressure had fallen to 50 mm. mercury. This was the conventional level chosen as a criterion early in our experience with shock before we came to appreciate that the attributes of shock usually develop long before this level is reached.

ADRENALIN SHOCK

The first studies were made on adrenalin shock. The normal volumes were determined and then the volumes in shock (table 1). Every case showed a reduction in blood volume; in two cases this reduction amounted to one-third of the total blood, in another case the blood volume was reduced by half. The blood counts on the other hand indicated a relatively small concentration, a finding that occasioned some surprise in view of the large concentrations obtained by Lamson (3) and by Bainbridge and Trevan (4). This discrepancy in results was traced directly to the acacia injected for the determination of the normal blood volume. One experiment was, therefore, performed in confirmation of the above authors. Four injections of adrenalin were made as indicated in figure 1*a*; with each of the first three there occurred

concentrations of the blood amounting to 29, 32 and 45 per cent of the normal respectively. In the intervals of lower blood pressure between the first three injections the red cell count returned to a condition in the neighborhood of normal. Each succeeding injection produced a concentration greater than the preceding although the adrenalin was given in smaller doses. Each injection must therefore have left a residuum of damage which became such after the third injection that the plasma did not return to the vessels as before. It

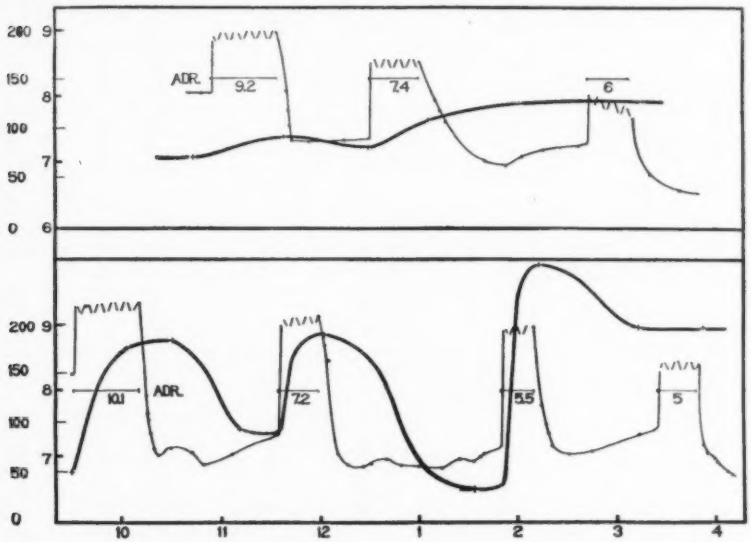

Fig. 1. (*a*) Lower: without acacia; (*b*) Upper: after the injection of 4 cc. per kilo of 20 per cent acacia. Light line: blood pressure in mm. Hg., indicated on the axis of ordinates. Heavy line: R. B. C. count in millions, indicated on the axis of ordinates. Axis of abscissas: time of day. Adrenalin injected at times indicated. Figures give numbers of cubic centimeters of 1/1000 injected.

is probable that only in this latter condition was the damage to the tissues sufficient to be spoken of as true shock.

A similar experiment was now performed in which 4 cc. per kilo of 20 per cent acacia were administered before adrenalin in comparable amounts was injected. The results (fig. 1*b*) show that the acacia not only has a marked effect on the final concentration attained but that it has an even more striking effect on the ebb and flow of plasma which

TABLE 1

Shock produced by intravenous injection of adrenalin

		R. B. C. COUNT, NORMAL	R. B. C. COUNT, SHOCK	PER CENT INCREASE	CORRESPONDING DECREASE IN BLOOD VOLUME, PER CENT OF NORMAL	CORRESPONDING DECREASE IN PLASMA VOLUME	NORMAL BLOOD VOLUME, PER CENT OF BODY WEIGHT	SHOCK BLOOD VOLUME, PER CENT OF BODY WEIGHT	PER CENT DECREASE	REMARKS
A. Without acacia	I	6,752,000	8,804,000	30.4 45.0 max.	23.4	39.0	9.5*	7.68	19.2	This experiment is a companion to the succeeding for comparison. Injection into the femoral vein
B. With acacia	I	7,096,000	7,956,000	12.0	10.7	17.8				
	II	4,520,000	4,630,000	2.4	2.35	3.9				In this and the succeeding 5 experiments adrenalin was injected into a mesenteric vein
	III	5,808,000	5,800,000	0	0	0	9.56	4.5	53.0	Heart stopped 30 minutes after injection for shock volume determination. Circulation continued 10 minutes by massage of the heart
	IV	5,856,000	6,216,000	6.16	5.82	9.7	10.7	7.25	32.2	Pressure 41 with regular heart. Typical shock tracing
	V	6,656,000	7,712,000	15.7	13.5	22.5	9.8	6.56	33.0	Adrenalin injection 5½ hours after first count; 60 cc. blood removed in samples. Shock typical

B. With acacia	VI	5,552,000	5,976,000	7.7	7.15	11.9				Irregular heart
	VII	3,496,000	3,624,000	3.6	3.5	5.8	9.9	9.2	7.0	Shock determinations 23 minutes after fourth injection of adrenalin. Pressure 22 mm. Hg.

* In this and subsequent tables the decrease in blood volume is figured as the reciprocal of the percentage of normal attained by the red blood cell count. The plasma decrease is estimated on the assumption that the plasma constitutes 60 per cent of the volume of the blood. In the last three columns are given the direct determinations of the blood volume by the acacia method. Where the normal volume is not determined it is assumed to be 9.7 per cent of the body weight, which is the average of the normal volumes of a long series of dogs.

result from the pressure changes, the concentrations being in this case 4, 8 and 11 per cent of the normal for the successive injections.

In experiment III of the acacia series a decrease in volume of 53 per cent by transudation alone would mean the loss of practically all the plasma. Obviously some other factors are involved in so large a depletion of the blood volume and of these, absolute stasis in some part of the vascular bed must be the most important. While the red cell count does not indicate any loss of plasma at all, this absence may be only apparent as the red cell count determines the minimum of concentration and the determination is misleading in proportion to the number of corpuscles that are jammed in the capillaries. In experiment IV, which was very satisfactory, a large difference between the absolute loss and the loss by concentration again occurs. That the difference is not merely apparent but may be due mainly to stasis, may be inferred after comparison with experiment VII in which, with a very small decrease in blood volume, such a degree of shock developed that the pressure was 22 mm. of mercury. Here undoubtedly the defective circulation must be attributed to stasis, which however falls short of being absolute.

In all but two of the experiments included in the table the adrenalin was injected into one of the small mesenteric veins. For this purpose care was taken to expose only one small loop of intestine. In these experiments curious localized constrictions of all branches of the mesenteric veins appeared which were permanent in position and gave the vein a sausage-like appearance. This phenomenon is probably one of the factors accounting for the loss of blood to the circulation by stasis.

SHOCK FROM CLAMPING THE AORTA

It was evident from the experience with adrenalin that in the other experimental procedures for producing shock the volume changes should be determined both without and with a previous injection of acacia. When no acacia had been injected clamping the aorta for three hours (table 2) produced a quite constant amount of concentration. As indicated by the red blood cell count the loss of plasma amounted to an average of 37.0 per cent of its original volume. On the contrary when acacia had been previously injected the plasma losses were 0 per cent and 8.8 per cent in two cases. Exactly similar results were obtained in the experiments in which the hemoglobin content was used as an index of the concentration (table 2 B., expers.

TABLE 2

TABLE 2

Shock produced by clamping the aorta

		R. B. C. COUNT, NORMAL	R. B. C. COUNT, SHOCK	PER CENT INCREASE	CORRESPONDING DECREASE IN BLOOD VOLUME, PER CENT OF NORMAL	CORRESPONDING DECREASE IN PLASMA VOLUME	NORMAL BLOOD VOLUME PER CENT OF BODY WEIGHT	SHOCK BLOOD VOLUME, PER CENT OF BODY WEIGHT	PER CENT DECREASE	CALCULATED DECREASE IN BLOOD VOLUME	REMARKS
A. Without acacia	I	6,150,000	8,048,000	30.8	23.7	39.5					
	II	7,624,000	9,464,000	24.1	19.4	32.3		8.4		13.4	Showed signs of improvement at end. Not in shock
	III	7,424,000	9,736,000	31.0	23.7	39.5					Blood volume not obtained on account of cardiac death. Voluminous mucohemorrhage exudate in intestine
	IV	7,608,000	9,768,000	28.3	22.0	36.7		7.9		18.5	Hemorrhage into intestinal mucosa. The count was taken before the clamp was removed from aorta
	V			17.1	14.6	24 4					From Hb. determinations
B. With acacia	I	6,240,000	6,104,000	0	0	0	9.54	5.65	40.8		Dog in profound shock when clamp removed
	II	7,568,000	7,992,000	5.6	5.3	8.8		6.7		31.0	No blood drawn before acacia was injected
	III						10.0	7.56	24.0		
	IV			3.0	2.9	4.8					From Hb. determinations
	V			2.7	2.63	4.4					From Hb. determinations

IV and V). Here again, as in shock produced by injection of adrenalin, the large differences between the actual decrease in volume and the apparent decrease due to loss of plasma, amounting respectively in two experiments to 40.8 per cent and 20.7 per cent, point to absolute stasis as the principal factor in decreasing the effective volume in these animals which had received acacia. In figure 2 are plotted the red cell counts obtained in two experiments performed for the purpose of comparing transudation under as similar conditions as possible except for the preliminary acacia injection in the one case. The concentration in the case without acacia was more than four times that in the control.

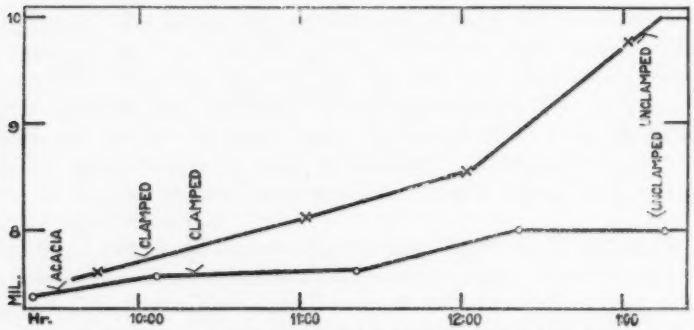

Fig. 2. R. B. C. count in shock produced by partial occlusion of the aorta. In the experiment indicated by the lower line 4 cc. per kilo of 20 per cent acacia were injected at the point designated.

SHOCK FROM MANIPULATION OF THE INTESTINES

In both of the two control cases (table 3), exposure of the intestines produced large concentrations of the blood. In these experiments the transudation of plasma could be observed directly on the peritoneal surface of the intestines. Beads of plasma would appear, grow larger, coalesce and start to run from the surface. In one experiment in which the drippings were collected, about 150 cc. were obtained. At the period of maximum transudation fluid was lost at the rate of 1 cc. per minute. This same phenomenon to a less degree can be seen also where shock is produced by other methods. If the abdomen is carefully opened droplets of serum are often seen on the peritoneal surfaces, especially of the liver and spleen.

TABLE 3

Shock produced by exposure of the intestines

		R. B. C. COUNT, NORMAL	R. B. C. COUNT, SHOCK	PER CENT INCREASE	CORRESPONDING DECREASE IN BLOOD VOLUME, PER CENT OF NORMAL	CORRESPONDING DECREASE IN PLASMA VOLUME	SHOCK BLOOD VOLUME, PER CENT OF BODY WEIGHT	CALCULATED DECREASE IN BLOOD VOLUME	REMARKS
A. Without acacia	I	6,664,000	8,944,000	34.2	25.4	42.6	5.8	40.0	Probable stasis in intestinal vessels
	II	5,192,000	6,904,000	33.0	24.7	41.1	7.5	22.7	Loss mainly by concentration
B. With acacia	I	8,168,000	8,504,000	4.1	3.95	6.6		32.0*	
	II	5,200,000	5,920,000	13.8	12.1	20.2	9.1	10.0†	
	III	5,536,000	7,488,000	35.0	26.0	43.4			

* Approximation.

† Calculated on volume obtaining after acacia injected without previous drawing of samples.

In this form of shock the difference between the experiments with and without acacia is not so marked. This may be due in the greater part to the fact that the damage produced can not be well controlled. The rapidity of the onset of shock in strong healthy animals varies with the severity of the manipulation. In one dog in the acacia series the concentration was maximal, in the other two the concentration amounted to less than one-half of that occurring in the controls. If we assume that the intestinal conditions are about the same in animals in which approximately the same time elapses between the exposure

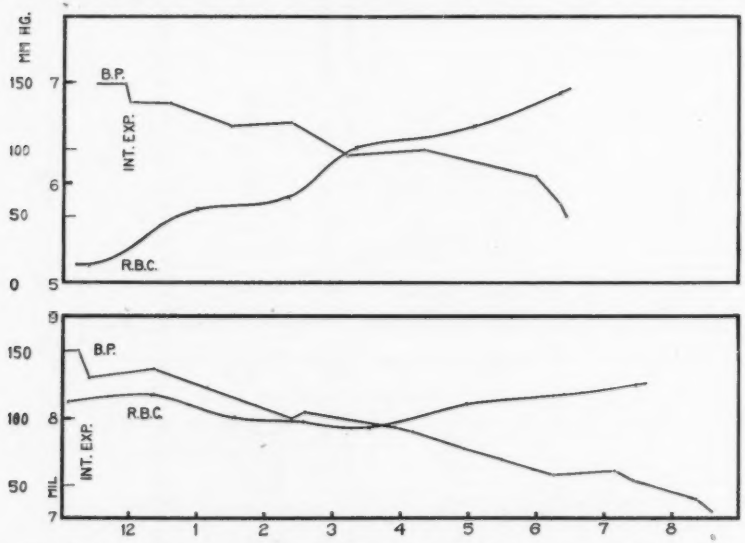

Fig. 3. Blood pressure and R. B. C. count in shock from manipulation of the intestines. Upper: without acacia. Lower: after the injection of 4 cc. per kilo of 20 per cent acacia.

of the intestines and the intervention of shock, then comparison of two such cases (fig. 3) shows that there is a considerably longer interval before the plasma loss becomes appreciable if acacia has been injected previous to the intestinal manipulation.

SHOCK FROM CLAMPING THE VENA CAVA

Four cases are included in this series (table 4). In all a concentration of the blood took place, the degree being somewhat greater in the two animals unprotected by acacia.

TABLE 4

Shock produced by clamping the vena cava

		R. B. C. COUNT. NORMAL	R. B. C. COUNT. SHOCK	PER CENT INCREASE	CORRESPONDING DECREASE IN BLOOD VOLUME, PER CENT OF NORMAL	CORRESPONDING DECREASE IN PLASMA VOLUME	NORMAL BLOOD VOLUME, PER CENT OF BODY WEIGHT	SHOCK BLOOD VOLUME, PER CENT OF BODY WEIGHT	PER CENT DECREASE	CALCULATED DECREASE IN BLOOD VOLUME	REMARKS
A. Without acacia	I	6,816,000	8,456,000	24.0	19.8	33.0		8.4		13.4	Pressure 70 mm. Hg., animal improving. Volume measured ½ hour after last count ( ) = Maximum. Organs especially hemorrhagic (Autopsy). Volume determination corresponds to smaller concentration
	II	7,908,000	8,448,000 (9,592,000)	6.8 (21.3)	6.3 (17.6)	10.5 (29.3)		7.4		23.7	
B. With acacia	I	6,456,000	6,608,000	2.3	2.3	3.8	9.8	9.0	9.2		
	II	6,400,000	7,220,000	12.8	11.3	18.8		8.5		16.0†	

† Calculated on volume after acacia injection.

In the second experiment in series A, the concentration of the blood was progressive until the clamp was removed. After this the red cell count decreased. At the end of the experiment the blood volume as determined directly was 23.7 per cent less than normal. If the circulation had been recovering as the cell count might indicate the pressure would not have progressively fallen and the final volume would not have been so low. A more probable explanation is that some of the corpuscles were jammed in the capillaries and therefore caused a decrease in the count, a view which is supported by the hemorrhagic condition of the organs at autopsy. In another series of nine animals in which the cava was clamped for 2 to 2½ hours so that the general blood pressure was 40 mm. of mercury, the concentrations at the end of clamping as indicated by hemoglobin determinations averaged 117.6 per cent of the normal, the individual variations ranging from 110.4 to 131.8 per cent.

DISCUSSION

In every case studied shock was associated with a concentration of the blood and loss of volume. This was not a phenomenon of severe shock alone, but started soon after the procedure was begun, the purpose of which was the production of shock. The concentrations did not always vary parallel to the loss in absolute volume.

In the slightly larger portion of the total number of experiments the total loss of volume can in the main be explained by the amount of plasma loss. Examination of the tables shows that in a few cases the shock volumes as determined directly are not quite small enough to coincide with the volume calculated from the red cell count. These differences are without significance as they are obviously errors in procedure and technique and the reasons for their occurrence must vary with the individual cases. The following possibilities may be mentioned: 1, In one case the direct determination was made one-half hour after the red cell count, in an animal that was recovering. 2, When the volume of the blood in shock is determined without a previous determination of the normal volume, the percentage decrease is calculated on the assumption that the normal volume was 9.7 per cent of the body weight, although in some cases it might have been actually higher than the average for normal dogs. 3, It is probable that if we had followed the hemoglobin content instead of the cell count, many of the differences would have disappeared as it has been our experience that the variations in volume calculated from the

erythrocyte count were almost uniformly greater than those calculated from simultaneous hemoglobin determinations. Scott, Herrmann and Snell (5) had a similar experience in their studies of the concentration of the blood after muscular exercise. These authors give a discussion of this point with a citation of the literature.

In the other group, cases can be found in which the total loss of blood as determined by the acacia method is much greater than that indicated by the red cell count. The greatest variations between the total loss as actually determined and that deduced from the concentration were seen in the adrenalin cases in which the injections were made into a mesenteric vein. In this condition the venous constrictions mentioned above appear as a special factor. In three cases where the total losses were 53, 32 and 33 per cent of the total blood respectively, the losses by plasma filtration were 0, 6 and 13 per cent. There can be no doubt that this special condition can only be incidental and does not even account for the differences found in this particular type of shock. As has been already mentioned, in two cases after clamping the aorta there were differences of 40 per cent and 20 per cent of the blood volume between the total loss and the loss by transudation. Similar cases also occur in shock from clamping the cava or manipulating the intestine, although the differences seen are not so great.

Before discussing the manner in which this blood is lost, it is necessary to review the post-mortem findings. In over one hundred animals examined it was found that no matter how the shock was produced the same general picture obtained. The most constant finding is an injection of the intestinal mucosa. In the milder cases the injection is confined to the duodenum; in more severe cases it extends along the whole of the small intestine giving it the appearance of deep red velvet. In the lower ileum the injection often again disappears although in severe cases it extends up to the ileo-cecal valve. In 60 per cent of the cases blood was found in the intestinal lumen; this was at times due to isolated punctate hemorrhages, more often a general sloughing of the tips of the villi took place. Microscopic sections show that except at injured tips of the villi the corpuscles are almost entirely within the vessels. The capillaries and small veins are greatly dilated and tightly packed with red blood cells. It is interesting to note that in animals that recover from this condition the injection has disappeared. How much this is due to sloughing and how much to clearing out of the capillaries we are unable to say. The spleen is usually swollen and has dark raised areas which consist of hemorrhages into the pulp. This

swelling and hemorrhage in some cases produces a spleen many times the normal size. The liver is occasionally found to be hemorrhagic on section.

Where the loss of blood volume as directly determined is much greater than that determined by the red cell count, the difference can be attributed mainly to stasis. The red cell count is an index only to loss of plasma, as a result of which concentration of corpuscles takes place. As long as no other factor enters, the blood volume can be properly interpreted in this way. But as soon as the corpuscles reach a certain concentration, 60 per cent according to Trevan (6), they become contiguous, the internal friction rises rapidly and they have a tendency to jam especially when the arterial pressure is decreased as it is in these experiments. Insofar as the corpuscles are blocking the capillaries they are not included in the determinations of the red cells or of the hemoglobin content, and the latter fail as indices of blood out of circulation by the volume of the corpuscles and the volume of the plasma that would normally accompany them. In addition to corpuscular sedimentation the difference may be attributable to sequestration of both corpuscles and plasma and to blood lost into the intestinal lumen and to a much less extent to hemorrhages into the spleen pulp.

In the group in which the total blood loss is to be accounted for by transudation, the congestion of the capillaries and the small veins is again seen post mortem; in these dilated and congested areas the blood must therefore be moving, however slowly.

The blood volumes found in severe shock are often remarkably high. Typical shock was found to occur in animals whose absolute volumes were decreased by only 7 to 17 per cent of the normal. For this there can be only one interpretation and that is an enlargement of the vascular bed and, therefore, a greatly reduced effective volume. Mann (7) in his experiments found that in shock from exposure of the intestines a portion of the blood becomes immoblized. On the assumption that the blood volume is 7.7 per cent of the body weight, he found that when all the blood that could be obtained was drawn from the femoral artery and heart, 24 per cent of the blood remained in the tissues of the normal dog while under similar conditions 61 per cent of the blood remained in the tissues of the dog in shock. We have often observed a similar phenomenon. From a dog in shock weighing 16 kilos, which would normally contain about 1400 cc. of blood, only 100 cc. could be obtained although artificial respiration and massage of the heart were resorted to. The decreased amount obtainable by bleeding is

in part due to the decreased volume, but analysis of Mann's data shows clearly that after allowance for the decreased volume of the blood in shock the actual amount of blood left in the body is more than in the normal dog. These findings are in line with the observations that have been reported by surgeons that fatal consequences may supervene on the losses of apparently trivial amounts of blood. The experimental animals maintain their pressure in the face of decreasing blood volume to what may be called a breaking point beyond which the pressure rapidly fails and death soon results.

In spite of the fact that the amount of blood which may be obtained by bleeding is so small (i.e., that so little of it can be moved from the stagnant area by the remaining vis a tergo after hemorrhage is started), it is still possible for the shocked animal to gradually move the mass of blood in the stagnant area so that an injected substance, as acacia, is gradually mixed with the total circulating blood. The figures for the blood volumes represent all the blood that passes through the heart in a period of ten minutes, and as indicated in the preceding paragraph, may amount to a total not far below normal. While the evidence indicates that the deficiency of the circulation is mainly due to sluggishness of the flow in some parts of the vascular bed, the work of Gesell (8) shows that a decrease of 7 to 17 per cent of the blood volume is itself of significance. He found that a loss of blood by hemorrhage of less than 10 per cent may elicit through constriction of central origin a decrease of flow through the submaxillary gland of more than 60 per cent.

After simple hemorrhage the volume would rapidly be made up from the tissues and the flow restored. In these experiments, on the other hand, the volume-compensating mechanism is becoming exhausted and the decrease in volume, though small, is sustained. If the constriction would be sustained, then the reduction in flow might cause damage to the parts of the body compensating the areas primarily injured. Robertson and Bock (9) have found that after a large hemorrhage in wounded soldiers the blood volume even when aided by transfusion may not return to normal for $2\frac{1}{2}$ days. The organism is, therefore, able to maintain its existence after large losses of blood without complete restoration of the volume for several days.

The locus in which the plasma is stagnant is certainly not the arterioles or portions of the vascular bed proximal to them. In the direct determinations of peripheral resistance to the inflow of salt solution (10) under a constant high pressure, it was found that while the periph-

eral tone was decreased when the pressure falls below 50 mm. Hg, in the periods preceding this low level the resistance may be but very little decreased, normal or even above normal. The condition of the large veins in the abdominal cavity can be easily noted by direct observation. Workers with experimental shock are agreed that in this condition the veins are collapsed and not engorged. There is left, then, only the capillaries and small veins. The full dilated capillaries and venules seen in the microscopic sections from the organs in shock prove their involvement beyond peradventure. It is interesting to note that Mall and Welch found that in the dog's mesentery after occlusion of the superior mesenteric artery the smaller veins become distended with corpuscles even before the capillaries. An opening up of normally unused capillaries such as occurs in inflammation may also well be a factor in enlargement of the capillary bed, as has been suggested by Cannon (11).

If one reviews the whole series of shock experiments and compares the concentrations obtained where shock was induced with and without an early injection of acacia, he finds that the average reduction of the blood volume by plasma loss was 6.8 per cent in the former case as compared with 20.3 per cent in the latter. It might be expected from such data that the animals that receive acacia would be more difficult to put into shock than their controls. There was, however, nothing observed in this series to contribute to this view. While on theoretical grounds it is to be expected that any benign method of interfering with transudation would be invaluable in the maintenance of normal blood volume, it should be fully appreciated that transudation is only one of the attendant circumstances in the development of the shock picture as a whole. That the transudation may be greater than indicated by the cell counts and may, therefore, alter the quantitative difference between the two series somewhat, should not be overweighted as the same phenomenon occurs in both series.

To explain the conserving action of the acacia on the plasma volume a consideration is necessary of the factors that might determine its loss. The loss of plasma might be due to a decreased colloidal content, to an increase of capillary pressure or to a change in permeability in the vascular endothelium.

The protein content of the plasma was studied in shock from clamping the aorta by determining its specific gravity with the pycnometer and its power of refraction with the Pulfrich refractometer. The latter method is theoretically more accurate as the salts have a relatively

greater specific gravity than protein but a smaller refractive index. The findings obtained with one instrument in the main confirm those obtained with the other. They show the same sequence of events in each case (table 5). There is in two experiments a slight increase of the protein content up to the time that the clamp is removed; in the other two experiments the increase is absent. Water evidently filters more rapidly than protein at first but after unclamping when the pressure is raised in the splanchnic region this is no longer true, the increased

TABLE 5

EXPERIMENT	BLOOD PRESSURES	HEMOGLOBIN PER CENT OF NORMAL	SPECIFIC GRAVITY OF SERUM	PER CENT OF PROTEIN IN SERUM (REFRACTOMETER)	REMARKS
I	105	100.0	1.0256	5.902	Normal
	58	112.4	1.0261	5.967	After 3 hours clamping
	40	113.3	1.0252	5.553	Final
II	105	100.0	1.0275	7.049	Normal
	58	111.4	1.0277	7.049	After 3 hours clamping
	40	111.4	1.0272	6.768	Final
III	180	100.0	1.0284	6.970	Normal
	100	114.1		7.416	After 3 hours clamping
	43	121.6		6.719	After second clamping
	43	122.5	1.0274	6.465	Final
IV*	100	100.0		7.675	Normal
	77	102.1		7.675	After 3 hours clamping
	30	101.6		7.653	Final

* This animal received 4 cc. per kilo of 20 per cent sodium acacia before the aorta was clamped.

permeability allows filtration of more protein and this increase is followed by a change in the opposite direction so that the protein content is finally 4 to 7 per cent less than at the start. It therefore follows that the plasma is lost mainly as a whole (see also Dale and Laidlaw (12)), and that a change in the colloid content of the plasma is not an important factor in the filtration as the latter process occurs during the period in which the colloid content is increasing as well as in the later period when the plasma is again diluting.

The explanation of the dilution in the final period probably lies in the fact that the organism is attempting to compensate the decreased

volume. The decrease in the protein content of the plasma would accord with the assumption that the passage of fluid from tissue to blood is by osmosis and therefore consists mainly of water and salts (13). As the loss of plasma is progressive all parts of the vascular bed are not similarly affected; in some portions the normal reaction to decreased volume is possible synchronously with a condition allowing continuous filtration in other portions. The compensation of the loss of fluid by normal tissues was first noted by Cobbett and Roy (14). Our data give the additional information that some areas are still compensating even when the blood is concentrating, though with insufficient rapidity to keep pace with the filtration.

The filtration of the plasma as a whole precludes the possibility that one finds suggested in the shock literature that the plasma is depleted by an increased affinity of the tissues for water.

In many cases a rise of capillary pressure must be a factor. After adrenalin the observed (15) rise of portal pressure must determine a rise in the mean capillary pressure of the intestinal area. When the cava is clamped there is a slight rise of portal pressure while the venous pressure in the liver which is normally very low must be relatively greatly increased. When the aorta is clamped it is difficult to see where in the splanchnic area the capillary pressure can be increased unless it be in capillaries plugged at their distal ends. Aside from this possibility transudation in this condition must be due to a change in vascular permeability. This latter condition is seen most clearly when the intestines are manipulated. Here, to be sure, alterations in pressure may also occur from various mechanical possibilities but the important changes are those which also occur in the inflammatory process, the analogy to which has been noted by a number of observers.

Regardless of the cause of shock, it would be expected that the expansion of the blood volume, which would result from the effort of the organism to compensate the increase of colloidal osmotic pressure of the blood produced by the injected acacia, would aid in maintaining the volume. To test the degree to which this might take place, data on the osmotic pressures developed by acacia and blood serum are necessary.

Osmotic pressures of acacia and blood serum. As Bayliss has correctly pointed out the determinations should be made against Ringer's solution. The two variables, acidity and CO_2 content of the solution, must be controlled. A difficulty also arises in the fact that acacia is altered by the presence of dilute sodium bicarbonate solutions. Natu-

ral acacia is combined mainly with calcium. Of the other constituents magnesium, another divalent cation, predominates. When acacia is added to a dilute sodium bicarbonate solution, some of the divalent cations are replaced by sodium, as is shown by the development of a precipitate. This takes place slowly if made up in the cold, more rapidly if warmed. We have found that sodium acacia develops twice the osmotic pressure that natural acacia does. This is true whether determined directly against water or by cryoscopy of concentrated aqueous solutions.

The osmotic pressure will depend on the nature of the cations with which it is combined when it comes to equilibrium in the blood. While we have not determined the quantitative effect of all the variables, we believe that we obtained a fair approximation of the natural conditions by dialyzing the acacia against Locke's solution without glucose but containing enough sodium bicarbonate so that when in equilibrium with a partial pressure of 40 mm. of CO_2 the reaction was about P_H 7.4. The acacia was made up to a 7 per cent strength in the above solution at room temperature. It was dialyzed in a Moore and Roaf osmometer (celloidin membrane) against the buffer mixture, arrangement being made for a continuous change of fluid in the lower chamber. Under these conditions the osmotic pressure of the 7 per cent solution was found to be 22 mm. of mercury.

Sodium acacia similarly prepared developed a pressure of 28.7 mm. of mercury.

These figures are evidently lower than those obtained by Bayliss (16). The difference is probably to be attributed to the differences in the other substances existing in the solution. A wide range of pressures may be obtained according toe the conditions of solution; for example, 6 per cent sodium acacia developed a pressure of 274 mm. of mercury in two days against CO_2-free distilled water; the water in the lower chamber was then changed to water saturated with CO_2 at atmospheric pressure and the osmotic pressure of the acacia fell to 83 mm. by the end of two weeks.

The osmotic pressures obtained with fresh dog serum dialyzed against the buffer mixture were also found to be lower than those usually reported, being 16.4 mm. of mercury as compared with 25 to 30 mm. for serum dialyzed against its ultrafiltrate (13), 27 mm. for pig serum against 0.9 per cent saline (17), and 36 to 40 for ox serum against Ringer's solution (16).

Theoretical considerations as to the effect of the injected acacia on the osmotic properties of the plasma. On the basis that the osmotic pressure of 7 per cent acacia is 22 mm. of mercury and of the protein of the blood serum 16.4 mm., 4 cc. of 20 per cent acacia would have to expand to $\frac{(20 \times 22 \times 4)}{7 \times 16.4}$ or 15.3 cc. to become isotonic to the serum colloids. This would mean a 16.6 per cent expansion of the blood volume (normal = 92 cc. per kilo). When 4 cc. of 20 per cent acacia were added to 54 cc. of serum and dialyzed against the same serum, the mixture developed a pressure of 3 mm. of mercury, being, therefore, 18.3 per cent hypertonic, and in experimental agreement with the calculation made from the osmotic pressures of serum and acacia determined separately.

The normal volumes and red cell counts were determined ten minutes after the acacia was injected. At this time the expansion of the blood is never more than 2 to 3 per cent above the calculated simple dilution by the volume injected. If we subtract this 6.4 per cent (4.4 per cent expansion by the volume injected + 2 per cent expansion due to dilution) from the 16.6 per cent increase in volume theoretically possible after the injection, there remains a further possible expansion of 10.2 per cent of the blood volume. The water which theoretically might be attracted by the acacia might be expected to account in part for the differences in plasma volumes in shock induced with and without aprevious injection of acacia, but there is no evidence that the filtration is masked by a progressive dilution of the injected acacia. In the normal dog after the injection of 4 cc. per kilo of 20 per cent acacia, the expansion of the blood is always less after one hour than it is after ten minutes, not more. The maximum actual expansion never reaches anything like the theoretical. This point is of considerable interest as it minimizes the importance of what would seem to be one of the most obvious explanations of the protective action of acacia, namely, its increase in volume by the attraction of water.

When the procedure whose purpose is the induction of shock is not started until one hour after the acacia injection, the conserving action of the acacia on the plasma volume is just as apparent. At the end of one hour as the blood has not expanded by absorption from the tissues and as direct chemical determinations have shown that at this time the blood contains about 92 per cent of the amount present at the start, the blood colloids must have an osmotic pressure greater

than normal (calculated[1] to be 17 per cent greater). This fact gives a clue to its probable action.

When the capillary filtration pressure is high it is opposed by the increased colloidal osmotic pressure as is strikingly seen in the adrenalin experiments. In conditions of increased permeability, other conditions being equal, the increase of filtration would again be opposed by the increased pressure of the colloids. This would be the case in the experiments where shock was produced by clamping the aorta. In cases where the injury to the vessels is sufficient acacia is apparently without beneficial effect. This is seen in experiment III in the shock series from manipulation of the intestines. We have also had a similar experience in some experiments with pulmonary edema resulting from the inhalation of irritating substances.

While the expansion of the blood is slight compared with what is theoretically possible, the blood plasma is potentially more able to take fluid from the tissues should such a demand arise. Evidence has been presented that the decreasing volume of the blood resulting from trauma is partially compensated by fluid from uninjured tissue. This process would be aided by the osmotic pressure of the acacia in the plasma and much more fluid would have to be absorbed from the tissues before the concentration of the blood colloids would go below normal.

Acacia contains calcium in ionizable form as it may be precipitated by oxalates. That calcium is combined with a large organic element and is not present as a salt impurity is indicated by the fact that the acacia can not be freed of the calcium by dialysis. The possibility that the inhibitory action on permeability might be due to calcium has been considered. If the calcium were the sole factor concerned the action would not be shared by sodium acacia. The latter, however, is not the case as sodium acacia has an undoubted protective action; in fact our most recent observations show that when either sodium or calcium

[1] Calculation.

55/59 × 16.4 = 15.28 (osmotic pressure of plasma when diluted with 4 cc. of water per kilo; plasma volume assumed to be 60 per cent of 92 cc. of blood per kilo).

4 cc. of 20 per cent acacia yields a 1.356 per cent solution in 59 cc. of water.

1.356/7 × 22 (osmotic pressure of a 7 per cent sol.) = 4.26.

4.26 × 0.92 = 3.92 (osmotic pressure of the acacia in the plasma 1 hour after injection).

15.28 + 3.92 = 19.2 (total osmotic pressure of plasma 1 hour after injection).

19.2/16.4 = 117, therefore the osmotic pressure of the plasma would be 17 per cent greater than normal.

acacia is dialyzed against Locke's solution containing sodium bicarbonate in equilibrium with alveolar air and with a reaction of P_H 7.4 that their osmotic pressures approach each other and presumably, therefore, when either form of acacia comes to chemical equilibrium with serum, the product is the same. Calcium action could arise, therefore, only from the salt resulting from the liberated ion.

SUMMARY

The blood volume was determined in shock by the method of Meek and Gasser and compared with the reduction in blood volume as determined by the enumeration of the red blood corpuscles.

The blood volume was found to be decreased in all forms of experimental shock studied and after all grades of damage.

Red cell counts or hemoglobin determinations are of value in indicating blood volume only when no absolute stasis occurs and when no corpuscles are jammed in the capillaries.

The effective volume of the blood may be reduced in the following ways:

1. By decrease in the volume of the blood as the result of:

a, Transudation of plasma.

b, Transudation of plasma and jamming of the corpuscles in the capillaries and venules, or the latter combined with

c, Absolute stasis in some part of the vascular system.

d, Hemorrhage into tissue, especially into the lumen of the intestines.

2. By dilatation of the capillaries and small veins with greatly decreased slowing of the circulation. This is always attended by some loss of plasma, but the latter may be relatively inconsiderable.

The transudation of plasma is greatly opposed by injection of 4 cc. per kilo of 20 per cent acacia before traumatization. The mechanism of action is believed to be due mainly to the antagonism to filtration by the resulting increase in the osmotic pressure of the plasma colloids. A discussion is given of the other possibilities.

BIBLIOGRAPHY

(1) Sherrington and Copeman: Journ. Physiol., 1893, xiv, 52.
Cobbett and Roy: Allbutt's System of medicine, 1897.
Vale: Med. Rec., 1904, lxvi, 325.
Mann: Surg., Gynec., Obst., 1915, xxi, 430.
Dale and Laidlaw: Brit. Med. Journ., 1911, i, 380.
(2) Meek and Gasser: This Journal, 1918, xlvii, 302.
(3) Lamson: Journ. Pharm. Exper. Therap., 1915, vii, 169.
(4) Bainbridge and Trevan: Brit. Med. Journ., 1917, i, 382.
(5) Scott, Herrmann and Snell: This Journal, 1917, xliv, 313.
(6) Trevan: Biochem. Journ., 1918, xii, 60.
(7) Mann: Loc. cit.
(8) Gesell: This Journal, 1919, xlvii, 468.
(9) Robertson and Bock: Journ. Exper. Med., 1919, xxix, 139.
(10) Erlanger, Gesell, Gasser and Elliot: Journ. Amer. Med. Assoc., 1917, lxix, 2009.
(11) Cannon: Journ. Amer. Med. Assoc., 1918, lxx, 611.
(12) Dale and Laidlaw: Loc. cit.
(13) Starling: Fluids of the body, 1909, 88.
(14) Cobbett and Roy: Loc. cit.
(15) Erlanger and Gasser: This Journal, 1919, xlix, 345.
(16) Bayliss: Proc. Roy. Soc., Ser. B., 1916, lxxxix, 380.
(17) Moore and Roaf: Biochem. Journ., 1907, ii, 34.

THE CATALASES OF THE BLOOD DURING ANESTHESIA

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There are two important theories in explanation of the mechanism by which anesthetics produce anesthesia. The one states that they produce their effects by diminishing the permeability of cell membranes (1). The other theory, that of Verworn and his followers (2), states that anesthesia is produced by an inability of cells to use oxygen. This inability is ascribed to depression of oxygen "activators" or oxygen enzymes. Our interest centered about these theories, and was renewed by publications in which it was said that the catalases of the blood were diminished in anesthesia. Added to this was the statement that this discovery forged another link in the chain of reasoning of the Verworn school (3).

Catalases may be defined as substances, if substances they are, which break up peroxides into molecular and thus inactive oxygen, and water. The function of catalases, according to most views, is to destroy peroxides when their presence would be harmful in the organism. Others subscribe to this effect, plus certain specific ones, such as the release of oxygen from hemoglobin. Von Furth however, who reviews the evidence, sums up by saying that "we not only know nothing positive about the physiological operation of the catalases, but also that we do not even know whether they have any important physiological significance at all, and whether they may not perhaps be altogether accidental and non-essential" (4). Burge (5), however, who with his associates has published numerous articles on this subject, claims great power and physiological significance for these catalases. The immediate ones which interest us are that they are decreased in anesthesia, according to his results, and that, since they are decreased, they are at least one of the causes for the decrease of oxidation in anesthesia, and hence for anesthesia itself. The interpretations and far-reaching conclusions which have been deduced from his results of catalase determinations under various conditions have seemed, *a priori*, too all-

conclusive. Recently F. C. Becht (6) has so ably and justly criticised not only his interpretations, but also his analytic data and methods, that it is unnecessary to repeat specific considerations here. Suffice it to say that we have met with the same difficulties in technique and in obtaining consistent results which Becht speaks of. The errors encountered in using these methods will be obvious to any one after a few trials. Our technique has not been as elaborate as that of Becht, but more so than that described by Burge. Blood was collected from an arm vein before anesthesia and immediately after operation was completed and the anesthetic stopped. Oxalate was used to prevent coagulation. The patients were unselected and were operated for the usual surgical diseases in the clinic of Dr. John B. Deaver. All determinations were made within a very short time after the collection of blood; 0.5 cc. of blood was added to 100 cc. of distilled water at 37°C. plus or minus 2 . The vessel containing this was immersed in a water bath at 40°, plus or minus 2°. Fifty centimeters of hydrogen peroxide were then added through a glass stoppered thistle tube passing into the bottle containing the blood and water. The evolved oxygen was collected through a second tube in an inverted burette. The apparatus containing the blood, water and hydrogen peroxide was shaken in a machine at a constant rate of one hundred and seventy-five double shakes per minute. The amount of oxygen evolved, first in 10 minutes, in later experiments in 5 minutes, was taken, reduced to 0°C. and 760 mm. pressure, as the measure of the catalases of the blood. The results in the table are typical of two hundred determinations, many done in duplicate, some in triplicate, and others done as many as four or five times. The same sample of hydrogen peroxide was used for the same individual before and after anesthesia, and as many constant conditions such as temperature, shaking, etc., were introduced as possible. Even so, to say nothing of the blood from different individuals, the blood from the same individual in duplicate or triplicate determinations gave results frequently differing by 50 per cent, and even 100 per cent, differences as large as in determinations before and after anesthesia. Of the whole number in which the most consistent results were obtained, 35 per cent showed an increase of catalases and 65 per cent showed a decrease. Granting that the analytic data are accurate, since the catalases were not universally diminished, they cannot, after all, play such an important rôle in anesthesia. Since, however, these methods of determining catalases are so highly inaccurate and varying, conclusions drawn from such observations can have no weight in physiological deductions.

TABLE 1

Catalases of the blood, before and after anesthesia. Cubic centimeters of oxygen evolved

BEFORE	AFTER	BEFORE	AFTER
316	381	418	353
417	410	430	412
92	32	250	379
372	414	55	179
841	1116	370	357
35	13	935	890

SUMMARY

Estimations of the catalases of the blood have been made before and after anesthesia. They were decreased in 65 per cent of the cases and increased in 35 per cent of cases.

The method for the determination of catalases has been criticised and the opinion expressed that they are inaccurate, and that no deductions can be drawn from them. Our results before and after anesthesia come within the experimental error (7).

The functions of catalases in the body are unknown.

BIBLIOGRAPHY

(1) For discussion, see LILLIE: Biol. Bull., 1916, xxx, 311.

(2) VERWORN: Bull. Johns Hopkins Hosp., 1912, xxiii, 97.

(3) BURGE, NEILL AND ASHMAN: This Journal, 1917, xlv, 388.

(4) VON FURTH: Chemistry of metabolism (transl. by A. J. Smith), Philadelphia, 562.

(5) BURGE ET AL: This Journal, 1916, xli, 153; 1917, xlii, 373; 1917, xliii, 58, etc.

(6) BECHT: This Journal, 1919, xlviii, 171.

(7) Results of previous determinations were published in Journ. Biol. Chem., 1918, xxxvi, 211, where no especial emphasis was laid on them. Our dissatisfaction with those results led to the present efforts, which were systematically and persistently persevered in, with the hope of securing something definite.

CONTRIBUTIONS TO THE PHYSIOLOGY OF THE STOMACH

LI. The Control of the Pylorus

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INTRODUCTION

From the earliest times anatomists and physiologists were so impressed with the "peculiar office and functions of the pylorus" that

> Van Helmont conceived it to be the peculiar seat of the soul, an opinion to which Willis gave a degree of support. Richerand ascribed to it something like intelligence, when he said that it has a peculiar tact which enables it to select from the contents of the stomach what is proper to pass through, while it rejects the remainder (1).

The more rational explanations which followed were essentially mechanical in nature but not entirely satisfactory. More satisfactory was the theory proposed by Cannon (2) that the pylorus was under the chemical control of the acid chyme. The wealth of conclusive experimental evidence in support of this theory leads early to its acceptance by clinicians and physiologists, so that at the present day its general correctness is not doubted. But Cannon (3) himself stated that other factors might modify the chemical control, especially under abnormal conditions. To this many observers will readily agree and will quickly point out that Cannon's theory not only fails to explain the emptying of the stomach in achylia gastrica but that Cannon never convincingly explained the rapid exit of H_2O and neutral egg white solution which left the stomach before the free acidity of the intragastric contents was present to effect an opening of the pylorus or sufficiently concentrated after ejection into the duodenum to effect its closure. More recently, Morse (4) reports a diminution in the rapidity of discharge of the stomach with an increase in the acidity of the gastric contents—quite opposed to the findings of Hedblom and Cannon (5)—but explains the discrepancy on the basis of difference in experimental method.

Spencer, Meyer, Rehfuss and Hawk (6) observed that 1.0 per cent sodium bicarbonate hastened the discharge from the normal human stomach. On the motor side, Carlson (7) reports an inhibition of hunger contractions by introduction into the stomach of both acid and alkaline solutions but that the inhibition is greater with a given concentration of hydrochlorie acid than with a corresponding solution of sodium bicarbonate. The experimental conditions of Cole on human beings quite closely approximate those of Cannon, made for the most part on the cat. Cole (8) finds that a meal of mixed foods and fluids begins to leave the stomach immediately after ingestion, "certainly before ingestion of a full meal is complete." Cole emphasizes here, as in a previous article (9), that the pylorus opens partly at each "systole" and that "during the stage of "diastole" when the gastric pressure is diminished, the sphincter may be closed to prevent the chyme from falling into the stomach." In short, he correlates the opening of the pylorus with the passage of peristaltic waves, and an increased tonicity of the stomach. Ortner (10) ascribes the opening of the pylorus to an optimum dilution of the gastric contents rather than to the presence of free acid in the antrum. Egan (11) corroborates Cole's findings that in health as well as disease the first portions of the food ingested by the fasting stomach may leave it at once.

The present short contribution likewise establishes an interdependence between the opening of the pyloric sphincter and the tonus rhythm of the stomach and emphasizes anew together with observations of other authors that certain motor activities of the stomach are intimately associated with the relaxation of that sphincter.

METHODS

The observations which led to this conclusion were obtained partly from fluoroscopic observations of the stomach of man (A. J. C.) following the ingestion of an emulsion of tragacanth carrying in suspension $BaSO_4$, while recording the motor activities of the stomach by the ballon method partly from observations on normal fasting dogs provided with a gastric and duodenal fistula. In the animals the duodenum was attached to the anterior abdominal wall about 1 to 2 cm. from the pylorus at the time of making an ordinary gastrostomy. Following the uneventful recovery from this double but simple operation, a small opening was made into the duodenum. Through the gastrostomy we introduced into the dog's stomach H_2O, milk, milk-

peptone solution, or a 2 per cent cooked starch suspension as such or after the addition of an amount of dog's gastric juice sufficient to show presence of free acid with a few drops of congo red solution. The balloon was next inserted through the gastric fistula and attached suitably to the manometer in the usual fashion (12). With the dog lying comfortably on its right side in the lap of an assistant we next introduced through the duodenostomy a piece of rubber tubing with opening directed toward the pylorus and with its tip no more than 1 cm. from that sphincter. Following a short period of temporary inhibition as a result of these preparatory manipulations, the dogs evinced a typical tonus rhythm. Each drop of fluid issuing from the tube in the upper part of the duodenum was registered immediately beneath the manometer tracing of the gastric activity by means of a signal magnet. Very often the drops issued in so rapid a succession that the record of the individual drops merged into a broad continuous band.

In man, the balloon was swallowed just prior to the ingestion of about 10 ounces of a drink of aqueous emulsion of tragacanth carrying in suspension $BaSO_4$. The subject was placed on the X-ray table, face down, and after attaching the rubber tubing from the balloon to the recording manometer, fluoroscopic observations were made on the passage of the test drink through the pylorus and into the duodenum. As soon as the duodenal cap filled and the dark mass quickly passed into the upper portion of the duodenum the fact was recorded by a signal magnet writing immediately below the writing point which was recording the gastric activities at the time and just prior to the opening of the pylorus.

RESULTS

The direct fluoroscopic observations in man showed plainly in confirmation of the contention of Cole (8) and others that the fluid contents were carried past the sphincter and into the duodenum just as soon as we could prepare the patient to record the fact. Peristaltic waves would pass over the stomach starting approximately in the middle of the body (13) of the stomach and course toward the pylorus. The pyloric sphincter would open and the dark mass would be hastily passed through the whole course of the duodenum. Figure 1 illustrates the record given during the period of such observations. It will be noted that the period of opening of the pylorus and the passage of fluid contents through the duodenum as recorded by the lowermost line is coincident with the record of tonus rhythm of the stomach which

by fluoroscopic observation consisted of plain peristalsis whose vigor, judging from the depth of the advancing ring of constriction, was considerably more powerful than the graphic record would lead one to suppose. This is probably due to the fact that the progressive increase in intragastric pressure resulting from the advancing peristaltic wave tends to be neutralized partly by the opening of the pyloric sphincter, partly by the relaxation of that part of the stomach wall over which it has just passed.

This tracing is strikingly similar to one taken by Rogers (14) and Hardt from man three hours after a dinner of beef steak, bread, butter,

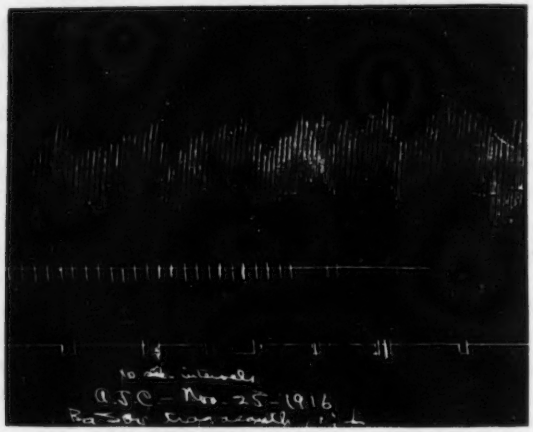

Fig. 1. A record of the movements of the stomach of man shortly after the ingestion of fluid carrying $BaSO_4$ in suspension. Lowermost line is a signal magnet tracing indicating the exit of the fluid contents of the stomach into the duodenum as determined with the fluoroscope. Middle line marks off ten second intervals.

apples and cream. The authors there refer to this motor activity as a "tonus rhythm." The experimental conditions (almost empty stomach) are the same as those here recorded. And since in our case the "tonus rhythm" was the graphic manifestation of peristalses coursing toward the pylorus we have no doubt that peristalses gave rise to their tonus rhythm if they had made fluoroscopic examination of the type of activity, particularly since the "tonus rhythm" is according to them gradually replaced as the stomach becomes empty by the typical

hunger contractions which are admitted by Rogers and Hardt to result essentially from peristalses starting at the cardiac orifice and passing "toward the pyloric end as a rapid peristaltic wave."

In short, the tonus contractions of the stomach accompanied by visible peristalsis (the "systole" of Cole) (15) effected an opening of the pyloric sphincter with egress of the gastric contents into the duodenum. In the dogs we recorded the drops of gastric contents issuing from a tube placed in the duodenostomy by a signal magnet writing just below the flag of the manometer recording stomach contractions after partly filling the stomach with H_2O, acidified starch paste or milk peptone mixture.

As soon as we had prepared the animal for the registration of the results, the stomach began to empty itself of its contents which were

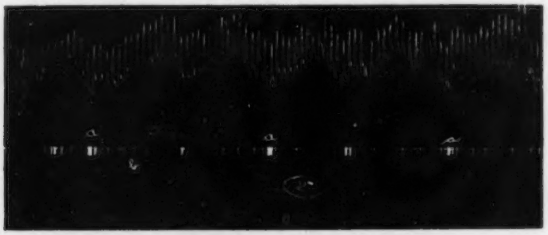

Fig. 2. Tracing showing the movements of the stomach containing food in a fluid condition. The signal magnet tracing below records the drops of fluid issuing from a duodenostomy located within an inch of the pylorus (dog).

often tinged with some bile. The fluid was acid towards phenolphthalein, rarely acid to congo red and dimethylamidoazobenzene. Though usually tinged with bile we could easily demonstrate the presence of starch and absence of sugar precluding the possibility of regurgitation of mixed intestinal contents (pancreatic juice, bile and succus entericus). It was, furthermore, never alkaline to phenolphthalein as noted above.

More interesting to us was the relation existing between the time of opening of the pyloric sphincter and the motor activity of the stomach. Figures 2, 3 and 4 are submitted to illustrate this relationship. Figure 2, obtained from a dog, is quite similar to figure 1 obtained from one of us (A. J. C.). In both instances the fluid is seen to issue during the rise in the intragastric pressure known to be due in the latter case to waves of constriction passing toward the pylorus (peristalses). As

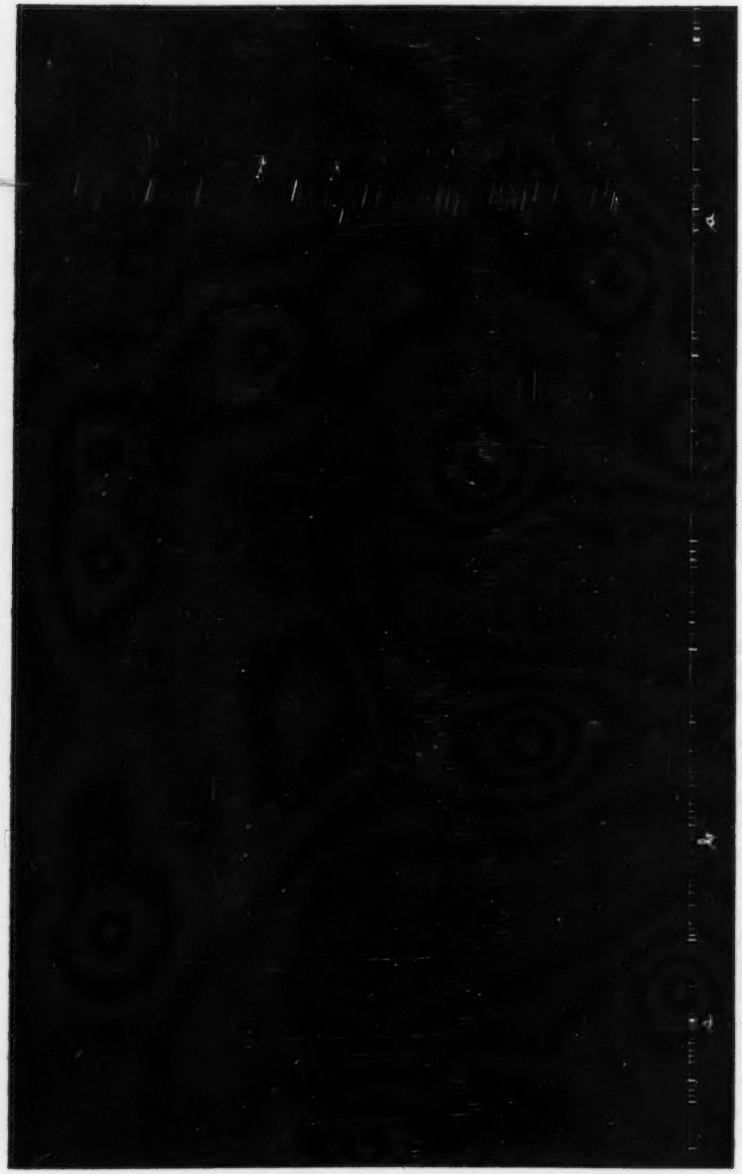

Fig. 3. Record of the movements of the partly-filled stomach with a signal magnet tracing below indicating the egress of the gastric contents through a duodenostomy located just below the pyloric sphincter.

the tracing plainly shows, the drops issued most abundantly at a time when the constricting ring had reached the pylorus. It may and does very often issue during the rise in tone of the stomach which may be due not entirely to a traveling ring of constriction (peristalsis) but to general increase in tonic activity of the musculature as a whole as is seen at *b* in figure 2, and better still at *b* in figure 3. Certain it is that the pylorus is most widely open, if judged by the quantity of intragastric contents issuing from the tube in the duodenum, at the

Fig. 4. Record of the movements of the partly-filled stomach with a signal magnet tracing below indicating the egress of the gastric contents in drops through a duodenostomy situated just below the pyloric sphincter.

time of or immediately after the rise in intragastric pressure as shown at *a* in figure 3 and in a particularly pure form at *a* in figure 4. In the last-mentioned case the contractions of the stomach which result in this ejection of chyme are identical with the contractions of the so-called empty or nearly empty stomach which are responsible for the sensation recognized universally as hunger pangs. Now, hunger contractions are known to result from peristalses sweeping over the

stomach toward the pylorus. We have, therefore, no hesitation in stating that there is certain relation between the opening of the pylorus and particularly vigorous peristalses but that a moderate general rise in intragastric pressure (fig. 3, *b*) in the absence of powerful peristalsis may effect the same result. But we again point out that the graphic record of the motor activity of the stomach is quite deceptive; for, where the activity was simultaneously observed fluoroscopically we have noted powerful waves of peristalsis when the record showed only moderate tonus changes (tonus rhythm of Rogers and Hardt). Comparing the contractions shown in figure 2, those of figure 4 are decidedly more vigorous. It is the latter type which is particularly related to an opening of the sphincter.

DISCUSSION

The results of Ivy (16) obtained after and quite independently of us are strikingly confirmatory of the observations just recorded. In describing emptying time of water (with ground meat) from the stomach, Ivy (p. 427) writes:

> In every dog it passed (i.e., the H_2O) in single gushes at varying intervals of from 10 to 30 seconds, each gush delivering from 5 to 30 cc. of water. These gushes as to time seemed to occur in groups, e.g., several of 10 second intervals would occur, then several of 15 seconds, then several of 12 seconds, and between these there might be interposed one or two of 5 second or of 30 second intervals. There was evidence of rhythm.

Although Ivy was not interested in the cause of this rhythm he concludes that "the gushes could easily correspond to the occurrence of the peristaltic waves or stomach contractions, as reported by von Mering in 1893." Ample evidence submitted by us in this paper justifies the assumption.

We are anxious to record here a difficulty which interfered for some time with a study of this problem. We planned originally to make direct observations, with the aid of a Kirstein light, of the behavior of the pyloric sphincter by looking at the sphincter through a glass cannula placed into the duodenostomy. Such attempts were invariably followed by pronounced and long-continued pylorospasm and vomiting. Vomiting is elicited with the greatest ease by any irritation of the duodenum. If great care is not exercised in introducing the collecting tube through the duodenostomy, vomiting invariably occurs. Reflex emesis is certainly more readily elicited from mechanical irri-

tation of the duodenal mucous membrane near the pylorus than from simple irritation of the gastric mucosa. Salivation and retching precede the act.

The failure of the fluid contents to reach the intestine under these conditions results from a true pylorospasm because of the duodenal irritation. Wave after wave of gastric peristalses would pass over the stomach but no ejection of chyme would follow. A silver director found the pylorus so tightly constricted that considerable pressure had to be applied before the instrument would pass into the stomach. Failure of fluid under these conditions to reach the intestine might have been due partly to a leveling of the gradient as conceived by Alvarez (17). Direct inspection, however, showed the pyloric sphincter tightly constricted (pylorospasm) and not patulous.

CONCLUSIONS

Sufficient evidence has been presented in this paper on the basis of experimental work on man and dog that there is a correlation between marked motor activity of the stomach (either as tonus changes or peristalses) and an inhibition in tone of the pyloric sphincter. The work of other investigators is not at variance with these results. On the contrary, most investigators recognize that factors other than the presence of free acid in the stomach near the pyloric sphincter are related to an opening of that sphincter.

Under the especial conditions described in our experiments, the intragastric contents issuing from the duodenostomy were usually acid toward phenolphthalein but rarely showed presence of free acidity to dimethylamidoazobenzene or congo red. Ivy (16) similarly found the water to issue neutral from the stomach. Only later did the acidity rise. Under normal conditions, according to Alvarez (18), the stomach wall follows gradients of irritability, rhythmicity and latent period from cardia to pylorus. With a gradient higher in the body of the stomach than near the pylorus, the increased pressure coming from above effects an opening of the pylorus even before the free acidity has reached a concentration sufficient to assume chemical control of that sphincter. It seems probable that even under normal conditions the chemical control has been greatly over-emphasized to the exclusion of other possibilities.

SUMMARY

1. Fluoroscopic examination of the stomach of man while recording simultaneously graphically the motor activity of the stomach shows that the pylorus opens for the ejection of chyme with arrival at the pylorus of powerful advancing rings of constriction aided possibly by a general increase in tone of the stomach musculature as a whole.

2. In dogs, the intragastric contents issue from a duodenostomy either *a*, during a marked rise in the intragastric pressure probably unaccompanied by peristaltic activity, or *b*, more commonly just at or after the peristaltic wave or waves passing over the stomach have effected their greatest increase in intragastric pressure.

3. There was certainly a greater relation between the muscular activity and the opening of the pylorus than between the latter and the reaction of the intragastric contents.

4. Vomiting is more easily induced by irritating duodenal mucosa than by an irritation of the gastric mucosa.

BIBLIOGRAPHY

(1) Quoted from BOSTOCK: An elementary system of physiology, London, 1836, 545.
(2) CANNON: This Journal, 1907, xx, 283.
(3) CANNON: Amer. Journ. Med. Sci., 1906, cxxxi, 569. For literature see ALVAREZ: Journ. Amer. Med. Assoc., 1915, lxv, 388.
(4) MORSE: This Journal, 1916, xli, 439; Arch. Int. Med., 1918, xxi, 48.
(5) HEDBLOM AND CANNON: Amer. Journ. Med. Sci., 1909, cxxxviii, 517. CANNON: The mechanical factors of digestion, New York, 1911, 120.
(6) SPENCER, MEYER, REHFUSS AND HAWK: This Journal, 1916, xxxix, 462.
(7) CARLSON: This Journal, 1913, xxxii, 245.
(8) COLE: This Journal, 1917, xliii, 618. To the best of our knowledge Cole did not publish his results in full. An inquiry addressed to him failed to elicit a response due to the fact, no doubt, that Doctor Cole is in the medical service of the army.
(9) COLE: Journ. Amer. Med. Assoc., lxi, 762.
(10) ORTNER: Pflüger's Arch., 1917, clxviii, 124. (Quoted from Physiol. Abs).
(11) EGAN: Berlin. klin. Wochenschr., 1917, liv, 506. (Quoted from Physiol. Abs).
(12) See figure in CARLSON: The control of hunger in health and disease, figure 4, 35.
(13) CANNON: The mechanical factors in digestion, 46.
(14) ROGERS AND HARDT: This Journal, 1915, xxxviii, 276, fig. 2.
(15) COLE: Journ. Amer. Med. Assoc., 1913, lxi, 762; This Journal, 1917, xlii, 618.
(16) IVY: This Journal, 1918, xlvi, 420.
(17) ALVAREZ: Journ. Amer. Med. Assoc., 1917, lxix, 2018.
(18) ALVAREZ: This Journal, 1916, xl, 585; 1917, xlii, 435.

PHYSIOLOGICAL STUDIES ON PLANARIA

II. Oxygen Consumption in Relation to Regeneration

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The present paper deals with the rate of oxygen consumption of Planaria during the process of regeneration. The physiology of this process has already been investigated in this laboratory by other methods and certain conclusions regarding the metabolic rate during regeneration have been reached. Chief among these methods is the direct susceptibility method, which consists, as explained in greater detail in the preceding paper of this series (1), in observing the time of death of living materials in certain toxic solutions. We believe for reasons previously given (1) that the time of death in these solutions is a measure of metabolic rate. By means of this direct susceptibility method, Child (2) has already determined the following facts regarding the metabolic rate of Planaria during regeneration.

1. The susceptibility of pieces of Planaria within the first few hours after they are cut is greater than that of intact worms similar to those from which the pieces came. Hence the rate of oxygen consumption should be greater after section.

2. The susceptibility of the main portion of the pieces then gradually falls and reaches a minimum in about twelve hours after section. From this time through three to four days after section it is about like that of intact worms. However, the susceptibility of the cut surfaces remains higher than that of the rest of the piece during this period and hence the total oxygen consumption should also be higher than that of intact worms. However, the factor of starvation must be considered, since, as shown in the first paper (1), the oxygen consumption decreases during the early stages of starvation.

3. The susceptibility of the pieces (both old tissue and regenerating surfaces) then begins to rise and continues to rise until the end of the regeneration process. Completely regenerated pieces are thus much

more susceptible than the original intact worms. The oxygen consumption should likewise increase during regeneration and the oxygen consumption of the regenerated pieces should be considerably greater than that of the original intact worms.

All of these expectations have been completely realized in the experiments to be reported. At the same time in this laboratory, Miss Harriet Robbins has determined the carbon dioxide production during regeneration and has found that it runs parallel to the oxygen consumption and the general susceptibility.

The present experiments were performed upon twelve different lots of *Planaria dorotocephala*. Each lot was selected at random from the general laboratory stocks and consisted of medium sized worms, 15 to 20 mm. in length, although no effort was made to select individuals of the same size, as this was not essential for the purpose of the experiments. The heads were removed from the worms the day before the experiment was begun in order to eliminate movement. The oxygen consumption of each lot of decapitated worms was determined. The worms were then cut into small pieces and the oxygen consumption of these pieces again determined at various intervals after section until the process of regeneration was completed. After each such determination the worms were weighed, and the oxygen consumption per unit weight could then be calculated. In some cases the regenerated worms were fed in order to eliminate the factor of starvation. In all cases, after regeneration was complete, the regenerated parts were cut off and the oxygen consumption of those portions which corresponded to the original pieces tested separately.

The method of determining the rate of oxygen consumption and the method of weighing have been described in the previous communication (1), to which the reader is referred for details.

Details regarding each lot of worms and the data obtained upon them are given in the following description and accompanying tables. To save space, the data upon lots 1 to 6 are condensed in table 1, only the final calculations of the rate of oxygen consumption being presented. The data upon lots 7 to 12 are given in detail in tables 2 to 7. In these last mentioned tables, the time since section is stated in the first column, the actual cubic centimeters of oxygen consumed in a given time interval in the second column, the weight in grams in the third, and the oxygen consumed per unit weight per unit time in the fourth column. For the purposes of this calculation, purely arbitrary units of time and weight were selected, units, however, approximating those actually

encountered in the experiments. The time unit selected is two hours, and the unit of weight 0.5 gram; and the figures given in all of the columns of table 1 and in the fourth columns of tables 2 to 7 are therefore the cubic centimeters of oxygen consumed in two hours by 0.5 gram of Planaria.

TABLE 1

Record of lots 1 to 6. Temperature 22°C.

CONDITION OF WORMS	OXYGEN CONSUMED BY 0.5 GRAM IN 2 HOURS			CONDITION OF WORMS	OXYGEN CONSUMED BY 0.5 GRAM IN 2 HOURS		
	Lot 1	Lot 2	Lot 3		Lot 4	Lot 5	Lot 6
	cc.	*cc.*	*cc.*		*cc.*	*cc.*	*cc.*
Decapitated, intact, 2 days since feeding	0 30	0.29	0.28	Decapitated, intact, 3 days since feeding	0.26	0.27	0.26
Cut into small pieces				Cut into small pieces			
No to 8 hours after section	0.35	0.34	0.32	No to 3 hours after section	0.32	0.35	0.31
	0.35	0.33	0.29				
	0.34	0.36	0.30				
1 day after section	0.35	0.31	0.33	2 days after section	0.33	0.32	0.36
4 days after section	0.27	0.27	0.29	7 days after section	0.30	0.30	0.32
8 days after section	0.32	0.28	0.31	13 days after section	0.32	0.32	0.38
16 days after section	0.54	0.55	0.53	22 days after section	0.45	0.41	0.43
Worms fed four times				Regenerated parts cut off			
23 days after section, 2 days since feeding	0.62	0.56	0.60	23 days after 1st section; few to 22 hours after second section	0.36	0.37	0.37
Regenerated parts cut off							
25 days since 1st section; 2 days since second section	0.59	0.53	0.63				

Record of lots 1 to 3 (table 1). The worms in these lots were collected on March 12, 1919; they were last fed on March 24; their heads were removed on March 25, and on March 26, the oxygen consumption of the decapitated worms was measured. It ranged in the three lots from 0.28 to 0.30 cc. of oxygen in two hours per

TABLE 2

Record of lot 7. Temperature 21°C.

CONDITION OF WORMS	OXYGEN CONSUMED IN TEST	WEIGHT	CALCULATION, AVERAGE AMOUNT OF OXYGEN CONSUMED BY 0.5 GRAM IN 2 HOURS
	cc.	*grams*	*cc.*
Decapitated, intact, 1 day since feeding	0.29 in 1½ hours 0.34 in 1½ hours*	0.799	0.26
Cut into small pieces			
1 to 8 hours after section	0.35 in 1½ hours 0.40 in 1½ hours	0.762	0.32
20 to 22 hours after section	0.39 in 1½ hours	0.673	0.38
2 days after section	0.36 in 1½ hours 0.35 in 1½ hours	0.628	0.37
7 days after section	0.20 in 1½ hours 0.23 in 1½ hours	0.501	0.28
15 days after section	0.32 in 2 hours 0.28 in 2 hours	0.363	0.41
Worms fed three times			
22 days after section; 1 day since feeding	0.39 in 2 hours 0.38 in 2 hours	0.419	0.45
Regenerated parts cut off			
23 days since first section; several hours since second section	0.26 in 2 hours 0.34 in 2 hours	0.308	0.48

* Whenever practicable, two separate determinations of the oxygen consumption were made on each occasion.

0.5 gram. The worms were then cut into small pieces and in several determinations taken during the first eight hours after section, the oxygen consumed ranged from 0.29 to 0.36 cc.—a distinct increase over the preceding figure. On the following day the oxygen consumption was about the same —0.31 to 0.35 cc. On March 31, four days after section, the oxygen consumption had fallen to 0.27 to

0.29 cc. On April 4, eight days after section, it was rising again, being 0.28 to 0.31; and on April 12, sixteen days after section, all of the pieces having undergone complete regeneration, the oxygen consumption was found to have increased greatly, having risen to 0.53 to 0.55 cc. The regenerated worms were now fed four times, on April 12, 13, 15 and 17, and on April 19 (the worms thus being in

TABLE 3

Record of lot 8. Temperature 21°C.

CONDITION OF WORMS	OXYGEN CONSUMED IN TEST	WEIGHT	CALCULATION, AVERAGE AMOUNT OF OXYGEN CONSUMED IN 2 HOURS BY 0.5 GRAM
	cc.	*grams*	cc.
Decapitated, intact, one day since feeding	0.37 in 1½ hours 0.39 in 1½ hours	0.952	0.26
Cut into small pieces			
1 to 8 hours after section	0.40 in 1½ hours 0.46 in 1½ hours	0.914	0.31
20 to 22 hours after section	0.48 in 1½ hours	0.813	0.38
2 days after section	0.45 in 1½ hours 0.40 in 1½ hours	0.755	0.37
7 days after section	0.25 in 1½ hours 0.24 in 1½ hours	0.598	0.27
15 days after section	0.35 in 2 hours 0.33 in 2 hours	0.435	0.39
Worms fed three times			
22 days after section; 1 day since feeding	0.48 in 2 hours 0.46 in 2 hours	0.502	0.46
Regenerated parts cut off			
23 days since first section; several hours since second section	0.28 in 2 hours 0.33 in 2 hours	0.369	0.41

the same state regarding nutrition as the original lots), the oxygen consumption was found to be 0.56 to 0.62 cc., a slight increase. The regenerated tissue at both ends of the worms was now cut off and two days later, on April 21, the remaining portions, corresponding as nearly as possible to the original pieces, were tested. Their oxygen consumption was 0.53 to 0.63 cc. The temperature throughout the experiments was 22°C. ± 0.5.

Record of lots 4 to 6 (table 1). The worms in this lot were collected during the early winter of 1919, and were large and well-fed specimens. They were last fed on April 7, their heads cut off on April 9, and the decapitated worms tested on April 10. The oxygen consumption was 0.26 to 0.27 cc. They were then cut into small pieces and within the first few hours after section, their oxygen consumption was found to have risen to 0.31 to 0.35 cc. Forty-eight to seventy-two hours after section, it was about the same, 0.32 to 0.36 cc. On April 17, seven days after section, it had fallen slightly to 0.30 to 0.32 cc. On April 23, thirteen days after section, it was rising again, being 0.32 to 0.38 cc.; and on May 2, twenty-two days after section, when the pieces had undergone complete regeneration, the

TABLE 4

Record of lot 9. Temperature 21°C.

CONDITION OF WORMS	OXYGEN CONSUMED IN TEST	WEIGHT	CALCULATION, AVERAGE AMOUNT OF OXYGEN CONSUMED IN 2 HOURS BY 0.5 GRAM
	cc.	*grams*	*cc.*
Decapitated, intact, one day since feeding	0.33 in 1½ hours 0.35 in 1½ hours	0.838	0.27
Cut into small pieces			
½ to several hours after section	0.38 in 1½ hours 0.40 in 1½ hours	0.790	0.33
21 to 23 hours after section	0.45 in 1½ hours	0.714	0.42
2 days after section	0.39 in 1½ hours 0.38 in 1½ hours	0.658	0.39
7 days after section	0.21 in 1½ hours 0.24 in 1½ hours	0.530	0.28
15 days after section	0.32 in 2 hours 0.30 in 2 hours	0.375	0.41
Worms fed three times			
22 days after section; 1 day since feeding	0.41 in 2 hours 0.41 in 2 hours	0.448	0.47
Regenerated parts cut off			
23 days since first section; several hours since second section	0.36 in 2 hours 0.39 in 2 hours	0.338	0.55

oxygen consumption was much higher than at the beginning of the experiment, being 0.41 to 0.45 cc. The regenerated portions were now removed and the oxygen consumption of the remaining pieces, corresponding to the original pieces, was 0.36 to 0.37 cc. The temperature throughout these experiments was also 22°C. ± 0.5.

TABLE 5

Record of lot 10. Temperature 18°C.

CONDITION OF WORMS	OXYGEN CONSUMED IN TEST	WEIGHT	CALCULATION, AVERAGE AMOUNT OF OXYGEN CONSUMED IN 2 HOURS BY 0.5 GRAM
	cc.	*grams*	*cc.*
Decapitated, intact, four days since feeding	0.23 in 2 hours 0.21 in 2 hours	0.599	0.18
Cut into small pieces			
1½ to 3½ hours after section	0.29 in 2 hours	0.512	0.28
18 to 22 hours after section	0.29 in 2 hours 0.23 in 2 hours	0.469	0.27
2 days after section	0.23 in 2 hours 0.20 in 2 hours	0.452	0.23
4 days after section	0.18 in 2 hours	0.403	0.22
8 days after section	0.17 in 2 hours 0.17 in 2 hours	0.332	0.25
17 days after section	0.15 in 2 hours 0.16 in 2 hours	0.181	0.42
Regenerated parts cut off			
22 days since first section; few hours since second section	0.05 in 2 hours 0.06 in 2 hours	0.113	0.24

Record of lots 7 to 9 (tables 2 to 4). The stock from which these worms were taken was collected in January, 1919. They were last fed on April 23, their heads removed on the same day, and their rate of oxygen consumption tested on April 24. They were then cut into small pieces and their rate of oxygen consumption tested at various intervals after section, as in the preceding experiments. As before, the oxygen consumption was found to be greater during the first few hours

up through two days after section than it was in the same worms before section; it then fell, as shown by a measurement on May 1, seven days after section. From this time on, the oxygen consumption rose and on May 9, fifteen days after section, it was considerably higher than that of the original intact worms. The regenerated worms were then fed on May 10, 12 and 14, and the oxygen consump-

TABLE 6

Record of lot 11. Temperature 18°C.

CONDITION OF WORMS	OXYGEN CONSUMED IN TEST	WEIGHT	CALCULATION, AVERAGE AMOUNT OF OXYGEN CONSUMED IN 2 HOURS BY 0.5 GRAM
	cc.	*grams*	*cc.*
Decapitated, intact, four days since feeding	0.23 in 2 hours 0.24 in 2 hours	0.560	0.21
Cut into small pieces			
1 to 3 hours after section	0.29 in 2 hours	0.540	0.26
18 to 22 hours after section	0.29 in 2 hours 0.25 in 2 hours	0.503	0.26
2 days after section	0.25 in 2 hours 0.23 in 2 hours	0.476	0.25
4 days after section	0.20 in 2 hours	0.430	0.23
8 days after section	0.20 in 2 hours 0.19 in 2 hours	0.360	0.27
17 days after section	0.17 in 2 hours 0.15 in 2 hours	0.153	0.52
Regenerated parts cut off			
22 days since first section; few hours since second section	0.06 in 2 hours 0.05 in 2 hours	0.085	0.32

tion tested on May 15, one day after feeding, as was the case with the original lots. As before, feeding resulted in a distinct increase in the rate of oxygen consumption. On May 16, the regenerated tissue was removed, and the oxygen consumption of the remaining pieces, corresponding to the original pieces, was again tested. The temperature throughout was 21°C. ± 0.5.

Record of lots 10 to 12 (tables 5 to 7). The members of lots 10 and 11 came from a stock which had been for some time in the laboratory (date of collection not known). They were large, well-fed individuals. The worms in lot 12 came from a general mixed stock containing material which had been used for other experimental purposes. The three lots were last fed on May 2, their heads were removed

TABLE 7

Record of lot 12. Temperature 18°C.

CONDITION OF WORMS	OXYGEN CONSUMED IN TEST	WEIGHT	CALCULATION, AVERAGE AMOUNT OF OXYGEN CONSUMED IN 2 HOURS BY 0.5 GRAM
	cc.	*grams*	*cc.*
Decapitated, intact, four days since feeding	0.23 in 2 hours 0.24 in 2 hours	0.681	0.17
Cut into small pieces			
½ to 2½ hours after section	0.30 in 2 hours	0.615	0.24
18 to 22 hours after section	0.31 in 2 hours 0.22 in 2 hours	0.566	0.23
2 days after section	0.28 in 2 hours 0.27 in 2 hours	0.532	0.25
4 days after section	0.22 in 2 hours	0.499	0.22
8 days after section	0.21 in 2 hours 0.21 in 2 hours	0.422	0.24
17 days after section	0.18 in 2 hours 0.20 in 2 hours	0.263	0.36
Regenerated parts cut off			
22 days since first section; 1 day since second section	0.11 in 2 hours 0.10 in 2 hours	0.166	0.31

on May 5, and the oxygen consumption of the decapitated worms was tested on May 6. The worms were then cut into small pieces, and the oxygen consumption of the pieces tested at intervals after section as in the preceding experiments. The rate of oxygen consumption was again found to be greater after section than before, remaining high through two days after section, then falling, and finally rising as regeneration was completed. The regenerated parts were then removed,

and the rate of oxygen consumption of the parts corresponding to the original pieces tested. The temperature throughout these three experiments was 18°C. ± 0.5.

A word is required regarding the degree of movement of the worms during these experiments, since movement increases the rate of oxygen consumption. The decapitated whole worms with which the experiments are begun are always perfectly quiet, as are also the pieces cut from them during the first ten or twelve hours after section. Twenty-four hours after section and from this time on through the greater part of the regeneration process there is some slight movement among the pieces but this is not sufficient to affect the measurements of oxygen consumption. The completely regenerated worms, however, usually move about considerably, and the figures obtained at the end of the regeneration process are probably slightly too high on this account, although the amount of movement was reduced in most cases by shading the worms and placing them in the flasks in which they were to be tested some time before the test was carried out. In experiments 10 to 12 inclusive, movement was practically completely eliminated in all cases by testing the worms at a temperature of 18°C. Since the results in these three experiments do not differ from those of the other nine, it is reasonably certain that movement is never a significant factor in the general result. After the regenerated worms are fed, they remain perfectly quiet, and the pieces obtained by cutting off the regenerated regions are also motionless.

CONCLUSIONS AND DISCUSSION

The data upon twelve different lots of worms are in accord with each other, and justify the following conclusions:

1. The oxygen consumption per unit time per unit weight of a given lot of worms is greater after these worms have been cut into small pieces than it was while they were intact. Section therefore increases the rate of oxygen consumption. The same conclusion had already been reached in this laboratory by means of other methods. Thus Child (2) long ago observed that the susceptibility of newly-cut pieces of Planaria to cyanide and other toxic solutions is much greater than that of intact worms. This increase in susceptibility is greatest at the cut surfaces but involves the entire piece also, in the case of small pieces. In long pieces the increased susceptibility is observable only at the cut surfaces and adjacent regions. This demonstrates that the

increase in susceptibility and rate of oxygen consumption following section are due primarily to the injury of cutting and spread from the cut surfaces with a decrement to the remainder of the pieces. The phenomenon is indeed only one example of the general physiological fact that injury is a form of stimulation, expressing itself in an increased rate of respiratory exchange, increased production of metabolic products, and electrical negativity.

The increase in susceptibility after section has been observed in this laboratory not only in Planaria but in a variety of the lower organisms. I found it to be true for several species of small fresh-water annelids (3) and incidentally in the course of numerous investigations upon the susceptibility of the lower forms, we have invariably observed that injured places are more susceptible than adjacent uninjured regions. Scott (4) noted that the oxygen consumption of the sea-anemone *Sagartia* is increased after section.

Not only are the rate of oxygen consumption and the susceptibility to toxic solutions of pieces of Planaria increased after section but, as Child (2) has shown with the aid of the Tashiro biometer, the carbon dioxide production is likewise accelerated. Recently Miss Robbins has repeated and extended these observations using the phenolsulphonephthalein method of Haas (5). Tashiro in his book *A Chemical Sign of Life* has shown that such an acceleration of carbon dioxide production as a consequence of injury is a practically universal biological phenomenon and he suggests that the occurrence of this change may be regarded as a proof that the material in question is living.

2. The increased rate of oxygen consumption following section was observed in these experiments to continue in most of the lots through forty-eight hours after section. Tests by the susceptibility method show that the susceptibility of the middle portions of the piece gradually falls after section and has returned to the condition found in intact worms in about twelve hours. However, the susceptibility of the cut and regenerating surfaces always remains higher than that of the middle part of the pieces. Hence the oxygen consumption, since it includes all parts of the pieces, must be greater than that of intact worms during this early part of the regeneration period. Nevertheless, it was expected that the oxygen consumption twenty-four and forty-eight hours after section would be somewhat less than it was within the first few hours after section. In lots 2, 5 and 10 this was the case but not in the other nine lots. It is therefore evident that extraneous factors enter into the determinations during this period, and this is further

indicated by the greater variability in the figures obtained during the first forty-eight hours after section than at later periods. It seems highly probable that the frequent weighings and handling during this period acted as sources of stimulation. This is further rendered probable by the greater uniformity in the measurements obtained with lots 10 to 12 which were tested at a lower temperature in order to eliminate some of these factors.

3. During the period from three or four days to a week after section the oxygen consumption is falling. Since the susceptibility method shows no such fall in metabolic rate during this time, the decrease must be regarded as due entirely to starvation. As shown in the first paper of this series (1), the rate of oxygen consumption decreases continuously during the first two weeks of starvation, this resulting from lack of activity of the digestive tract. In the case of regenerating pieces the total oxygen consumption does not decrease as a result of starvation to as great an extent as in non-regenerating worms, since this decrease is partially compensated by the increased metabolic rate of the regenerating ends of the pieces. The total oxygen consumption, therefore, of the pieces during the period from three or four days to a week after section is the resultant of two conditions, an increase due to regeneration and a decrease due to starvation. During this period the latter factor predominates.

4. From a week after section to the completion of the process of regeneration the rate of oxygen consumption is continually increasing. The oxygen consumption of the completely regenerated worms is very much greater than that of the intact worms from which the pieces were taken. The amount of this increase ranges in the twelve lots of worms from 50 to 150 per cent. Regeneration is thus a method of increasing the metabolic rate of Planaria. This conclusion had already been reached in this laboratory for Planaria and other forms through the use of the susceptibility method and further for Planaria through a study of carbon dioxide production during regeneration. Scott (4) found that the rate of oxygen consumption of a sea-anemone is increased by regeneration.

The objection may be raised that the increase in rate of oxygen consumption per unit weight during regeneration may be only apparent since the decrease in weight might be due to a loss of non-respiring materials. But if this were the case, the oxygen consumption should increase during the early stages of regeneration, when the weight is decreasing most rapidly. As a matter of fact however, both in the

present series of experiments and in the experiments on starvation previously reported, the oxygen consumption is also decreasing during this time. In the later stages of both regeneration and starvation, the loss of weight occurs at about a uniform rate or even decreases slightly; yet during this time the rate of oxygen consumption is continually rising. It is therefore reasonably certain that the observed acceleration of oxygen consumption in regeneration and starvation represents a real increase in the basic metabolism of the cells of the organism. The same conclusion was reached by Benedict (6) in his study of the metabolism of man during prolonged fasting.

The present experiments therefore support Child's conception of regeneration as a method of bringing about rejuvenescence—that is, restoring the organism to a metabolic condition comparable to that of young animals.

5. The regenerated worms of lots 1, 2, 3, 7, 8 and 9 were fed in order to bring them to a state of nutrition comparable to that of the original pieces, since, as already explained, starvation tends to lower the rate of oxygen consumption during the period in which the pieces are regenerating. In all six cases, the rate of oxygen consumption of the regenerated worms was increased by feeding. The tests were of course made at the same time interval after the last feeding as had been the case with the original pieces. It is naturally impossible to assert that after three or four feedings the regenerated pieces are in the same state of nutrition as the original pieces from which they came but at least they are as the result of such feeding more comparable to the latter. It is only after the effect of starvation has been eliminated that the real extent of the rise in oxygen consumption resulting from regeneration can be detected.

6. The rise in rate of oxygen consumption due to regeneration involves not only the newly regenerated portions, but also the old tissue of the piece. This was determined by removing and discarding the new heads and tails and testing the rate of oxygen consumption of the pieces remaining after this operation, such pieces corresponding as nearly as possible to the original pieces. Presumably these pieces are stimulated by section as were the original pieces although we cannot say whether they are stimulated to the same degree through the removal of the regenerated portions as are pieces cut from whole worms, as this point has not yet been subjected to experimental test. It is probable that they are less stimulated by section than are pieces cut from whole worms since in newly regenerated worms the old portions

have not yet established connections with the newly formed ends. Probably the severance of morphological and physiological connections and conduction paths is one of the chief factors in the stimulation observable after cutting.

Since, however, we have no definite information upon the degree of stimulation by section in pieces cut from regenerated worms, it is necessary to assume that it is the same as that in pieces cut from individuals not recently regenerating. One must therefore compare these pieces comprising the old portions of the regenerated worms with the original pieces at approximately the same lengths of time after section. The necessary data for this comparison are given in the tables. It is evident that when the regenerated worms were fed, the oxygen consumption of the old portions cut from them is always considerably higher than that of the original pieces the same length of time after section and after feeding. When the regenerated worms were not fed, the oxygen consumption of the old tissue is less but in all cases, except in lot 10, it is still higher than that of the original pieces considered the same length of time after section. This one exception may be due to variability in the degree of starvation.

It is therefore certain that when restored to the same condition of nutrition, pieces from which regenerated tissue has grown out have a higher rate of oxygen consumption than the same pieces before such growth occurred.

It is further evident from the data at hand that the rate of oxygen consumption of the old portions of the regenerated worms is less than that of the regenerated regions since in all cases where the worms were not fed, the oxygen consumption is reduced by cutting off the regenerated tissue. Where the worms were fed, a comparison cannot be made since the rate of oxygen consumption after feeding depends upon the number of worms which feed and upon the amount of food which they ingest, and this in return depends upon the morphological condition of the digestive tract, a factor which is very variable in a mixed lot of regenerating worms such as those used in these experiments.

SUMMARY

1. The oxygen consumption of a given lot of Planaria per unit weight is increased when they are cut into small pieces. This increase is due to the stimulation of injury.

2. This increase persists through about forty-eight hours after cutting but may fall slightly during this period. The persistence of the

increase is associated with the activity of the cut surfaces, the original tissue in the pieces probably not being involved.

3. The oxygen consumption of the pieces then falls and remains at a low level for about one week. This fall is due entirely to starvation. The fact, however, that the decrease in rate of oxygen consumption during this period is not as great as in starving non-regenerating worms indicates that the oxygen consumption of the regenerating regions has remained high throughout.

4. The oxygen consumption then begins to rise and continues to rise as regeneration proceeds. The oxygen consumption of the completely regenerated worms is 50 to 150 per cent greater than was that of the worms from which the pieces were taken.

5. When the regenerated worms are fed in order to eliminate the factor of starvation their oxygen consumption rises to a still higher figure.

6. The increased rate of oxygen consumption in regenerated worms is due not only to the high metabolic rate of the regenerated tissue but also in part to an acceleration of the rate of oxygen consumption of the old tissue comprising the original pieces. In other words, when a piece of Planaria undergoes regeneration, its metabolic rate is thereby accelerated; its rate is not, however, as high as that of the regenerated regions.

7. These experiments confirm the conclusion already reached by Child through other methods that the process of regeneration is a rejuvenating process, restoring the organism to a metabolic condition comparable to that of young organisms.

BIBLIOGRAPHY

(1) HYMAN: This Journal, 1919, xlix, 377.
(2) CHILD: Journ. Exper. Zoöl., 1914, xvi, 413.
(3) HYMAN: Journ. Exper. Zoöl., 1916, xx, 99.
(4) SCOTT: Proc. Soc. Exper. Biol. and Med., 1916, xiii, 121.
(5) HAAS: Sci., 1916, xliv, 105.
(6) BENEDICT: Carnegie Inst. Washington Publ., no. 203, 1915.

EFFECT OF ANESTHESIA AND OPERATION ON CERTAIN METABOLITES

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A previous study of anesthesia demonstrated certain changes in the acid-base equilibrium during that process (1). For practical needs in the matter, the question of whether to give carbon dioxide with the anesthetic or alkali before or after the anesthetic, was continually the basis of the work. A certain few surgical patients show uncompensated acidosis after anesthesia and operation. These studies were to show, if possible, changes in the nitrogen metabolism.

TECHNIQUE

Blood was collected before anesthesia was begun, and after the anesthetic was stopped, from the arm veins of unselected surgical patients admitted to the Lankenau Hospital, and operated in the clinic of Dr. John B. Deaver. Oxalate was used to prevent clotting. Blood-urea was determined by the urease method. Non-protein nitrogen was determined on 10 cc. of blood; coagulation of the proteins was accomplished by trichloracetic acid, and nitrogen estimated on the filtrate by the Kjeldahl method. Urine in the first series was collected twenty-four hours before, and twenty-four hours after operation; and in later experiments, it was collected at shorter intervals. The results were exactly the same. Urea was determined by the urease and titration method; ammonia by aeration and titration; phosphates by the uranium titration method; total acidity by titration with deci-normal sodium hydroxide, using phenolphthalein as indicator to the first faint pink.

Our a priori arguments were as follows: If acids are increased during anesthesia, the titrable acidity of the urine should increase. The blood-urea should diminish as a result of increased demand for ammonia. The urine urea should possibly decrease and the urine

ammonia increase. The figures shown in the table express the results of determinations on about ninety patients. The urinary acidity was increased very definitely in every case until in some instances, almost half normal acid was excreted. The blood-urea and non-protein nitrogen were increased in every case. In the urine the general tendency of the ammonia was to increase, whereas the excretion of urea was either increased or diminished, with apparently no relation between the urea in the urine and in the blood. The question of retention or increased production immediately suggested itself in the case of the increase in the blood-urea and non-protein nitrogen.

TABLE I

BLOOD				URINE											
Non-protein nitrogen per 100 cc.		Urea per 100 cc.		Ammonia				Urea				Titrable acidity			
				Before		After		Before		After		Before		After	
Before	After	Before	After												
mgm.	*mgm.*	*mgm.*	*mgm.*	*gm.*	*per cent*	*gm.*	*per cent*	*gm.*	*per cent*	*gm.*	*per cent*	*cc.*	*per cent*	*cc.*	*per cent*
40.4*	37.8	35	43	0.150	0.097	0.121	0.060	2.21	1.43	1.93	0.97	106.0	69	302	101
23.0	40.0	13	20	0.006	0.007	0.045	0.043	0.36	0.41	1.30	0.92	1.7	2	176	126
33.6*	21.5	13	22	0.073	0.048	0.039	0.060	1.62	1.06	0.47	0.73	79.0	55	87	134
42.8	44.5	24	34	0.166	0.070	0.365	0.121	3.27	1.39	5.17	1.72	42.0	18	498	166
44.2	48.1	22	27	0.026	0.052	0.047	0.047	0.43	0.87	0.57	0.57	29.0	58	80	80
29.9	34.4	12	16	0.022	0.019	0.041	0.045	1.45	1.25	0.72	0.80	19.0	17	83	93
36.9	43.6	11	30	0.020	0.081	0.268	0.063	0.59	1.74	7.35	1.73	14.0	40	408	115
44.2	49.2	13	38	0.232	0.082	0.016	0.055	3.96	1.41	0.35	1.19	112.0	40	3	110
35.8	48.1	12	15	0.084	0.030	0.240	0.092	4.11	1.46	3.40	1.41	47.0	17	325	125
39.4	44.8	25	45	0.053	0.070	0.094	0.049	0.66	0.88	0.89	0.47	16.0	22	83	44

* These were the only cases in the series which decreased.

MacNider's excellent work has shown that during anesthesia, in a non-nephropathic animal, the response to diuretics is approximately normal (2). From the fact that the blood-urea was universally increased, and the fact that among these patients there were numbers whose kidneys were non-nephropathic, as far as it was possible to learn, we can say that if retention does play a part, it does not tell the whole story. Therefore, there must have been an increased metabolism in the usual way down to urea. There must, however, also have been some slight relative changes in the nitrogen metabolism; for the increases in the non-protein nitrogen do not entirely parallel the increases

in blood-urea. As regards the urinary findings, the urea excretion bears no constant relationship to the variations in the blood-urea. This is in harmony with observations of urea excretion and blood-urea levels in other circumstances. We are not convinced that any definite and strict mathematical relationship has been shown as yet. That the ammonia is not increased in each case is perhaps not as surprising as first thought would suggest. A markedly increased elimination of phosphates accounts for some of the acidity of the urine (3). These no doubt play a part, as well as the sodium bicarbonate, in neutralizing the increased acids. In any given case, then, alkali is available in a number of forms, and in all probability, could a complete analysis be

TABLE 2

Urinary elimination of phosphates (P_2O_5)

BEFORE	AFTER
per cent	*per cent*
0.89	1.2
0.304	1.1
0.392	0.922
0.25	0.865
0.09	0.46
0.31	0.732

made of all the acids and alkalies, both in the blood and urine, before and after anesthesia, a mathematical proportion between them could be worked out.

The possibility has been kept in mind that the changes in metabolism may not be altogether along the normal route. Further investigations, particularly directed to a search for abnormal intermediate products, as well as, perhaps, the products of an entirely abnormal metabolism,—those due, for example, to direct destruction of cytoplasm or nuclear material by the anesthetics,—must be conducted to clear up some points. This was afield of our practical needs in the matter, and we have not carried them out.

SUMMARY

It has been demonstrated that small but very definite changes in metabolism take place during anesthesia. The reduction in bicarbonate of the blood plasma which occurs in that case must be due to

these changes, and not to mere over-ventilation, as suggested by Y. Henderson (4). Therefore, giving carbon dioxide with the anesthetic is contraindicated. Alkali should be given in selected cases.

BIBLIOGRAPHY

(1) Reimann and Bloom: Journ. Biol. Chem., 1918, xxxvi, 211.
(2) MacNider: Journ. Exper. Med., xxviii, 501, 517.
(3) Sawyer, Baumann and Stevens: Journ. Biol. Chem., 1918, xxxiii, 109.
(4) Henderson and Haggard: Journ. Biol. Chem., 1918, xxxiii, 333, 371.

EXPERIMENTAL SURGICAL SHOCK

V. The Treatment of the Condition of Low Blood Pressure which Follows Exposure of the Abdominal Viscera[1]

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This work was undertaken for the purpose of investigating under standard experimental conditions all the more important methods of treating a condition which exhibits the clinical signs of surgical shock in order to determine the relative value of the methods. While numerous such investigations have been made, only a portion of the entire field has been covered by a single investigation. Since in the different series of experiments various methods were used to produce the condition called shock it is impossible to compare the therapeutic results.

The method of producing the signs of shock was the same throughout in the present investigation and while the results may not be applied directly to all cases clinically diagnosed as surgical shock, the different methods of treating a condition which presents a common symptomatology can be accurately compared. Since it is obvious that the condition which the surgeon terms shock is due to a variety of causes, it is useless to attempt to find a specific therapeutic procedure, but as the symptoms are usually the same it is reasonable to suppose that some general therapeutic measures will be found.

The method of producing shock has varied greatly with the different investigations. In my work exposure of the abdominal viscera has seemed to afford the nearest approach to the production of shock presenting all the clinical signs (17). Our routine method was as follows: The animal, a dog, which had been fasted for from twelve

[1] Presented before the American Physiological Society, April, 1919, Baltimore.

to eighteen hours, was etherized in a closed cabinet, incubated, and a constant surgical anesthesia maintained by means of a Connell apparatus. Carotid blood pressure was recorded by means of a mercury manometer; sometimes the membrane manometer was also employed. After a record of normal blood pressure was obtained, the abdomen was opened and the viscera exposed. The only trauma to which the exposed viscera were subjected was the occasional gentle sponging with dry gauze or changing them from one side of the body to the other. When blood pressure had decreased and remained rather stationary at the desired level, which usually occurred about one to two hours after the exposure of the viscera, they were returned to the abdominal cavity and the wound was closed. After a length of time sufficient definitely to determine that blood pressure did not increase, procedures to improve the condition of the animal were instituted. The blood pressure was taken as a criterion of the animal's condition because it affords the easiest method of comparison. All other clinical signs of shock were also noted.

The maintenance of a constant anesthetic throughout the experiment removed the possibility of an error either in the interpretation of blood pressure records or in the general condition of the animal (19), (20). Anesthetic control experiments were carefully performed, the etherization being maintained at the same tension and for a length of time equal to that of the shock experiments. Practical conclusions can only be drawn from the results which apply to the condition in which the signs of shock were produced by exposure of the abdominal viscera, although it would seem that they should also be of value in a condition of a lowered and progressively decreasing blood pressure. The experiments were acute because it seemed that the therapeutic procedure would be put to a greater test if the animal was maintained under an anesthetic throughout the experiment and because the character of the experiment would not warrant the complete withdrawal of the anesthetic.

The fact should be emphasized that blood pressure which has been decreased and remains stationary below a certain level, and is allowed to remain there even for a very short time, is never restored and maintained by any known method of treatment. We have estimated the pressure below which no hope for restoration could be held as half the initial pressure maintained constant for one hour. In our experiments the methods of treatment, with very few exceptions, proved of no permanent value if the blood pressure had been decreased to less

Fig. 1. Kymograph record showing the successful use of acacia. *Record I*, normal blood pressure 118. *Record II*, after the abdominal viscera had been exposed 1½ hours; blood pressure 70. *Record III*, taken 30 minutes after the viscera were replaced; blood pressure 74; 160 cc. (20 cc. per kgm.) of a 6 per cent solution of acacia in 0.9 per cent sodium chlorid solution were injected (signal A-B); the injection was probably too rapid. The blood pressure increased to a maximum of 84. *Records IV, V, VI, VII* and *VIII* were taken at succeeding hours after the injection. The blood pressure was 84, 90, 95, 95 and 96 respectively. This is one of the few successful results following the use of acacia in the series of experiments.

Fig. 2. Kymograph record showing the favorable action of acacia. *Record I*, normal blood pressure 105. *Record II*, after 1 hour of exposure of the abdominal viscera; blood pressure 70. *Record III*, 15 minutes after replacing the viscera; blood pressure 68; 100 cc. (20 cc. per kgm.) of a 6 per cent acacia solution in 0.9 per cent sodium chlorid were injected slowly; blood pressure increased to a maximum of 82. *Record IV*, taken 30 minutes after injection; blood pressure 76. *Record V*, taken 1 hour after injection; blood pressure 80. *Records VI, VII, VIII* and *IX* were taken at successive hours after the injection with blood pressure of 92, 90, 98 and 85 respectively. These experiments seem to show that the result is as good as can be hoped for with acacia.

Fig. 3. Kymograph record showing the results of the injection of an alkaline acacia solution. *Record I*, normal blood pressure 148. *Record II*, after exposure of the abdominal viscera for 1 hour; blood pressure 88. *Record III*, after exposure of the abdominal viscera for 2 hours; blood pressure 75. *Record IV*, after replacing the viscera for 10 minutes and the injection of 146 cc. (20 cc. per kgm.) of a 7 per cent solution of acacia and 4 per cent solution sodium bicarbonate; the blood pressure increased to a maximum of 118. The succeeding records were taken at one-hour intervals after injection. The decrease in blood pressure occurring in the last records is characteristic of the action of an alkaline acacia solution, but usually blood pressure is not maintained for so long a time.

Fig. 4. Kymograph record showing a failure of acacia solution followed by a success with gelatine solution (Hogan's). *Record I*, normal blood pressure 140. *Record II*, 1½ hours after exposure of the abdominal viscera; blood pressure 82. *Record III*, 30 minutes after the replacing of the viscera; blood pressure 94; 104 cc. (20 cc. per kgm.) of a 6 per cent solution of acacia in 0.9 per cent sodium chlorid were injected (signal A-B). The injection may possibly have been made too rapidly; at any rate there was little improvement in blood pressure. *Record IV*, taken 35 minutes after the injection of acacia; blood pressure 70; 104 cc. of gelatine solution (Hogan's) were injected at the same rate the acacia had been injected; blood pressure increased to 112. *Records V, VI, VII* and *VIII* were taken at succeeding hours after the last injection. The blood pressure was respectively 100, 112, 118 and 110. The gelatine produced a much better result than acacia.

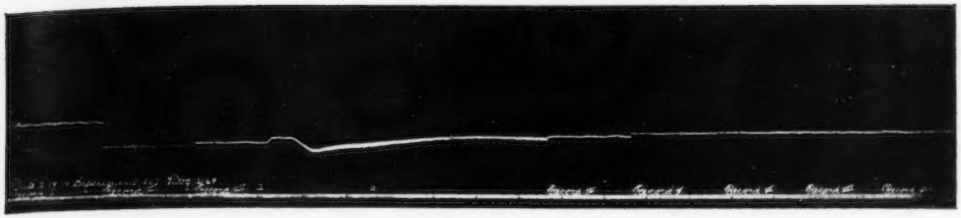

Fig. 1

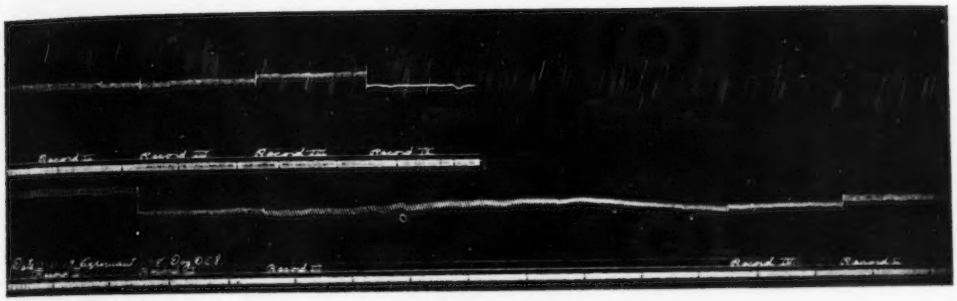

Fig. 2

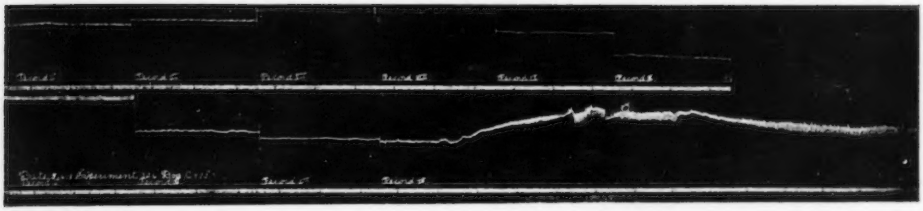

Fig. 3

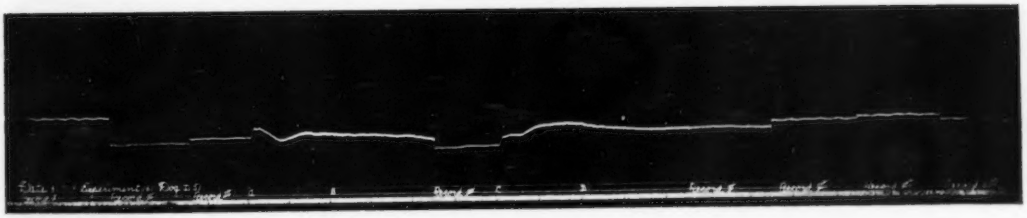

Fig. 4

Fig. 5. Kymograph record showing the action of the citrated blood after a failure of acacia and modified acacia solution. *Record I*, normal blood pressure 112. *Record II*, after exposure of the abdominal viscera for 1 hour; blood pressure 52. *Record III*, after the replacing of the viscera for 5 minutes; 70 cc. (20 cc. per kgm.) of a 6 per cent solution of acacia in 0.9 per cent sodium chlorid solution were injected (signal A-B). The blood pressure increased to a maximum of 80, but soon began to decrease and in 30 minutes it was 55. *Record IV*, taken 40 minutes after *record III*; blood pressure 50; 70 cc. of a modified acacia solution (6 per cent acacia, 10 per cent glucose, 1 per cent sodium carbonate, 1 per cent sodium sulphate were injected (signal C-D). The blood pressure increased to a maximum of 110, but soon began to decrease. *Record V*, taken 40 minutes after *record IV;* blood pressure 60. *Record VI*, taken 1 hour after the last injection; blood pressure 56; 70 cc. of citrated blood were injected (signal E-F). The blood pressure increased to a maximum of 85. *Records VII, VIII, IX* and *X* were taken at succeeding hours after the last injection. Note the beneficial action of blood.

Fig. 6. Kymograph record showing the restoration and maintenance of blood pressure by the injection of blood after a failure of acacia solution. *Record I*, normal blood pressure 105. *Record II*, 1 hour after exposure of the abdominal viscera; blood pressure 70. *Record III*, immediately after the replacing of the viscera; blood pressure 40; 70 cc. (20 cc. per kgm.) of a 6 per cent acacia solution in 0.9 per cent sodium chlorid solution were injected (signal A-B). The blood pressure increased to a maximum of 70, but within 30 minutes had decreased to 50. *Record IV*, taken 50 minutes after *record III;* blood pressure 48, 70 cc. citrated blood were injected (signal C-D). The blood pressure increased to a maximum of 86. *Records V, VI* and *VII* were taken at succeeding hours after the last injection, with blood pressures of 88, 90 and 88, respectively.

Fig. 7. Kymograph record showing the effect of injection of dextrin followed by gelatine solution. *Record I*, normal blood pressure 115. *Record II*, 1 hour after the exposure of the abdominal viscera; blood pressure 65. *Record III*, 15 minutes after the replacing of the viscera; 150 cc. (20 cc. per kgm.) of a 20 per cent dextrin solution were injected. The blood pressure increased to a maximum of 120, but soon decreased to 60. *Record IV*, taken 1 hour after *record III;* blood pressure 58; 150 cc. gelatine solution (Hogan's) were injected; blood pressure increased to a maximum of 82. The succeeding records were taken at one-hour intervals after injection. Note the slow recovery and the failure of blood pressure.

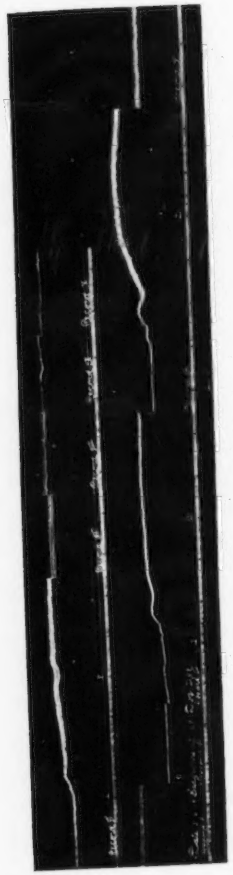

Fig. 5

Fig. 6

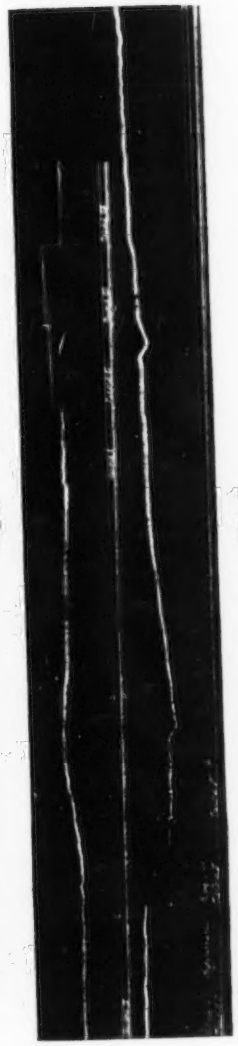

Fig. 7

Fig. 8. Kymograph record showing the results of the injection of dog serum. *Record I*, normal blood pressure 110. *Record II*, after exposure of the abdominal viscera for 1 hour; blood pressure 64. *Record III*, after the replacing of the viscera for 15 minutes; 110 cc. (20 cc. per kgm.) of dog serum were injected. The blood pressure was increased to a maximum of 110. The succeeding records, *IV* to *VIII*, were taken at one-hour intervals after injection. The serum restored blood pressure to normal and maintained it for 5 hours, until the experiment was interrupted.

Fig. 9. Kymograph record showing the beneficial action of dog serum after a failure of normal salt solution. *Record I*, normal blood pressure of 112. *Record II*, after exposure of the abdominal viscera for 1 hour; blood pressure 75. *Record III*, after the viscera had been replaced for 10 minutes; blood pressure 65; 156 cc. (20 cc. per kgm.) of normal salt solution were injected (signal A-B). The blood pressure increased to a maximum of 85, but in 15 minutes had decreased to 72. *Record IV*, an equal amount of dog serum was injected (signal C-D). Blood pressure increased to a maximum of 120. *Records V, VI, VII, VIII* and *IX*, were taken at succeeding hours after the injection, with blood pressures respectively of 105, 110, 120, 115 and 105.

Fig. 10. Kymograph record showing the beneficial results of citrated blood after a failure of acacia. *Record I*, normal blood pressure 130. *Record II*, after exposure of the abdominal viscera for 1 hour; blood pressure 80. *Record III*, after the replacing of the viscera for 15 minutes; blood pressure 70; 116 cc. (20 cc. per kgm.) of a 6 per cent acacia solution in 0.9 per cent sodium chlorid solution were injected slowly (signal A-B). The blood pressure increased to a maximum of 90, but soon began to decrease. *Record IV*, taken 30 minutes after the injection was stopped. Blood pressure 60. *Record V*, taken 1 hour after the injection; blood pressure 70. *Record VI*, taken 1½ hours after the injection; blood pressure 74; 116 cc. of citrated blood were injected slowly (signal C-D). The blood pressure increased to a maximum of 110 and then decreased slightly after the injection was stopped. *Record VII*, taken 30 minutes after the injection was stopped; blood pressure 118. *Record VIII*, taken 1½ hours after the injection; blood pressure 118. *Record IX*, taken 2½ hours after the injection; blood pressure 112. *Record X*, taken 4 hours after the injection; blood pressure 98. The animal was used for another experiment.

Fig. 8

Fig. 9

Fig. 10

than one-half its initial value; this is true regardless of the means by which blood pressure is lowered, for example, by hemorrhage, exposure of abdominal viscera, and by obstruction of the venous return, such as partial occlusion of the vena cava. Conclusions should, therefore, not be drawn with regard to a therapeutic procedure when it is tried out in an experiment in which the blood pressure has been decreased below one-half its normal value and because of the variability of the different animals unless, of course, the animal recovers. We believe however, that it might be of some clinical value if a therapeutic procedure could be found which completely or in greater part restores and maintains blood pressure for two hours, with the animal under a constant anesthesia and with a constant artificial temperature condition, after the blood pressure has been decreased from one-third to one-fourth its initial pressure by exposure of the abdominal viscera. These conditions were the standards we used in judging the value of the various methods of treatment.

The treatment of shock may be described under four headings: *a*, general measures; *b*, special measures; *c*, the use of drugs; *d*, attempts to restore fluid volume.

General measures. The most important general measure in the treatment of shock is the ancient practice of applying heat. The employment of heat is of value not only because shock is commonly associated with exposure to cold but also because the thermogenetic and thermo-regulatory mechanisms are impaired. It is probably not true that this impairment of the mechanism which keeps the body temperature constant is the primary cause of shock but the artificial maintenance of body temperature during the period of impairment produces beneficial results. It should be noted, however, that this deficiency in regulation applies to heat as well as to cold and that too much heat is harmful.

In most instances the temperature of our animals was kept almost constant by the judicious employment of an electric heating pad. In some experiments the heat was only applied after low blood pressure had been produced and at the same time the other therapeutic measures were instituted. Except that the blood pressure decreased more slowly when the heat was used from the beginning of the experiment no notable difference was observed in the results of the therapeutic procedure.

In order to increase the circulation around the bulbar centers it is usually recommended that the head be placed in slight Trendelenburg

position. Theoretically this should be of value, practically it may be; but in our experiments little effect could be noted.

Special measures. The purpose of most of the many special measures which have been devised for treating shock is to increase blood pressure either by decreasing the vascular capacity or by aiding in the return of blood to the heart. Strapping the limb and increasing intra-abdominal pressure should be of benefit, inasmuch as such measures decrease the vascular capacity, but their value is difficult to demonstrate experimentally. Rebreathing has also been recommended; according to Porter, it increases the action of the respiratory pump and thus aids the return of blood to the heart by sucking it into the thorax. The rationale of rebreathing in treating shock from the chemical standpoint is an integral part of Henderson's acapnial theory. The value and limitation of rebreathing in surgery and anesthesia were first carefully studied by Gatch (11). In previous studies on rebreathing in shock I have shown that the process is similar in the normal and in the shocked animal but that no measurable benefit results; this was confirmed by the present series of experiments.

Drugs. Drugs are usually employed in the treatment of shock for one of two purposes; first, either as a general stimulant, with particular reference to their action on the circulation and on the central nervous system (strychnin, camphorated oil, alcohol, etc.); and second, as vasomotor constrictors (epinephrin, pituitrin, etc.). Following the theory that shock is due to excessive vasoconstriction, the nitrites have been recommended for the purpose of decreasing vasoconstriction. Morphin is also recommended, mainly for its depressing action on the central nervous system.

Many investigations have been made with regard to the value of strychnin in the treatment of shock. Most investigators are agreed that the drug is of no value, although many surgeons, relying on their clinical experience, still use it in large doses (9). In experimental shock it is impossible to observe any effect of strychnin in doses smaller than those necessary to produce definite convulsive movements. It is questionable whether even these large doses produce a beneficial action; in our experiments it could not be said that any of the so-called stimulants were of value.

The value of the use of vasoconstrictors in the treatment of shock is still an open question. In the first place, although the decreased blood pressure is of great importance in shock, it is not known whether or not its increase by means of vasomotor constrictors is in itself of much

permanent benefit to the organism, and it would seem that they might be of distinct harm by decreasing the fluid supply to the tissues. In the second place, none of the vasoconstrictor drugs produce a very prolonged effect. Epinephrin is the most popular of these drugs to be employed in shock. It easily restores the decreased blood pressure and by continuous injection, the blood pressure can be maintained for a considerable length of time. As soon as the injections are stopped, however, the blood pressure sinks to its former level or usually lower. In our experiments pituitary extract produced a more prolonged action and seemed to be of somewhat greater benefit than epinephrin. Of course, repeated doses of the former cannot be employed as in the case of the latter drug. In general it may be said that experimentally the vasoconstrictor drugs produce little if any permanent benefit in the treatment of surgical shock although they might be employed clinically.

The nitrites produce their characteristic depression of blood pressure when it has been decreased by exposure of the abdominal viscera, but certainly no beneficial result has been observed from their use. Neither is the effect of morphin marked, but since it has been shown that the drug changes the regulations of blood volume, it should be studied more fully (2).

Attempts to restore fluid volume. It has been shown that a definite and marked loss of circulating fluid accompanies low blood pressure after the exposure of the abdominal viscera (18). This also seems to be true in other forms of experimental shock (12). In many clinical cases of surgical shock there is a loss of circulating fluid (4), (5), and it seems logical to treat the condition by an attempt to restore the lost fluid to the circulation. A large number of artificial fluids have been devised for this purpose. We have investigated the use of most of these solutions under the standardized conditions mentioned.

We usually injected the fluid to be tested with a burette although in some instances a continuous injection machine was employed. The former method proved the most practical, although it was impossible accurately to control the rate of injection. The effect of the injection depends somewhat on the rate at which the solution enters the vein. In general it seemed that the best results were produced with a rate that was just a little less than the amount which produced cardiac disturbance. Better results were obtained when the temperature of the solution was below 37° rather than above.

In our experiments the use of blood gave far better results than the use of any other substance except blood serum. If blood pressure is

not decreased to less than one-half its initial value after exposure of the abdominal viscera, the intravenous injection of citrated blood in relatively large amounts, 20 cc. per kgm., will practically always restore and maintain it for many hours. As a rule, equally good results have not been secured with any of the artificial solutions. Blood frequently restored blood pressure after other solutions had failed. Homologous blood serum will produce practically the same results (19).

Blood or blood serum show in many ways their superiority over all artificial solutions. They do not raise blood pressure more than some of the other solutions and quite frequently the blood returns more slowly to normal than after the use of some artificial mediums, but, whereas in most instances in which artificial mediums are used blood pressure soon drops to the shock level or below, after the injection of blood or serum, the increase in pressure is usually maintained for many hours. In shock the injection of any solution brings about a return of sensibility requiring higher ether tensions. The degree of sensibility is more marked after the injection of blood or serum than after any one of the solutions.

Physiologic sodium chlorid solution is usually employed to restore lost fluid volume; this is the least valuable of the artificial fluids used if the blood pressure has been lowered in the manner employed by us. Hypertonic saline solutions have been recommended and in some of our experiments they produced a definite beneficial action but the increased blood pressure was never long maintained. None of the saline solutions alone will maintain blood pressure for more than a very short time even when it has been reduced to but slightly below normal by exposure of the abdominal viscera. The saline solutions will usually pass out of the vascular system almost as fast as they are run in.

The use of sodium carbonate and bicarbonate in hemorrhage and shock was experimentally investigated several years ago; their use clinically has been emphasized recently.

Howell (16) seems to have been the first to study the effect of an alkaline salt in shock. He studied the effect of injection of sodium carbonate in a condition of shock produced by different methods. The beneficial results of such injections in the experimental conditions of low blood pressure which he had produced were due, he concluded, chiefly or entirely to a direct action on the heart. Dawson (6) in continuing Howell's study, investigated the effect of the injection of sodium bicarbonate in a condition of low blood pressure produced by hemorrhage. He found that it produced better results than the

sodium chlorid solution, and suggested that the bicarbonate solution be used in those cases of shock accompanied by hemorrhage. Seelig, Tierney and Rodenbaugh (23) obtained marked beneficial results by the injection of sodium carbonate in the condition of experimental shock, and concluded that the results were not due to the bulk of fluid injected, the hypertonicity or alkalinity of the fluid, or to the free carbon dioxid, but to the specific action of the salt on the heart muscle.

Cannon (4), in his study of shock in the front line trenches, found that there is a definite decrease in the alkalinity of the blood in cases of shock. The injection of sodium bicarbonate relieved this and produced very marked benefit. Patients on whom the surgeon refused to operate were tided over the critical period by the injection of either sodium carbonate or sodium bicarbonate which produced a rise in blood pressure and especially an increase in pulse pressure, thus making it possible to operate in a very short time.

In our experiments more lasting benefit was secured by the injection of sodium carbonate or sodium bicarbonate than by normal salt solution. Neither of the alkaline salts, however, completely restored blood pressure, nor was the increase long maintained.

Glucose has been suggested and used in post-operative treatment by several clinicians (3). Erlanger and Woodyatt (8) investigated its action in experimental shock and found it to be of some benefit. In our experiments the injection of such solutions was of definite value although rarely was there a complete restoration of blood pressure, nor was the increase long maintained. Glucose when added to some of the other artificial solutions seemed to enhance their value.

Hogan (15) first recommended gelatine as a medium to restore lost fluid volume. His formula was used in several of our experiments and gave good results in some. In general it is as satisfactory as any of the artificial mediums. Great care should be taken, however, in its preservation, because it deteriorates very readily and may produce untoward results. It was very difficult to modify the gelatine solutions by the addition of other substances, and no modification was found to be as safe or to give better results than Hogan's original formula.

We have used acacia and its various modifications as recommended by Bayliss (1). The addition of acacia to a transfusion solution certainly increases the power of that solution to restore and maintain blood pressure. The results following the use of acacia, however, were quite variable and sometimes disastrous. This variability of

action seemed to depend on both the acacia and the condition of the animal. It is quite possible that the results of our use of acacia have not been so good as those which others report because our acacia was not the same (21). We obtained the best we could, however, and I am quite sure the average surgeon who wishes to use it clinically would obtain no better. The alkaline acacia solution, when properly made by the addition of sodium carbonate or sodium bicarbonate, usually produced a better result than acacia alone, but it is difficult to prepare an alkaline acacia solution and more difficult to sterilize it and, on the whole, it did not seem to be a safe solution to use. Good results were produced by the addition of glucose to acacia (7). The modified acacia solution, which gave the best results in our experiments, consisted of 6 per cent acacia, 10 per cent glucose and 1 per cent sodium sulphate.

Many other methods for restoring fluid volume besides those already mentioned were tried. The rapid injection of 35 per cent solution of cane sugar, as recommended by Guthrie (13), usually fully restored the blood pressure but it was not long maintained. In acute hemorrhage, however, it produces good results. Various strengths of dextrine solutions were used; they restored the blood pressure more satisfactorily than the other artificial solutions but they failed to maintain it. A 1 per cent sodium sulphate solution produced fair results. It is interesting to note that distilled water gives better results in experimental shock with regard to blood pressure than do normal salt solutions. Crude preparations of hemoglobin from dog's blood produced good results and seems to warrant future study.

In summarizing the various methods employed to restore fluid volume it should be emphasized that *a*, in these experiments blood or blood serum produced by far the best results; *b*, the colloidal solutions were the best artificial solutions used; *c*, in general the gelatine solutions produced a more favorable action than the acacia solutions although some of the modifications of the acacia solutions produced as good or a better action than the gelatine; and *d*, care must be exercised in the use of gelatine and acacia because dangerous reactions may be produced with either.

SUMMARY

All the more important methods of treating under standard experimental conditions a state that exhibits the clinical signs of surgical shock which is produced by the exposure of the abdominal viscera of a dog, under a constant ether anesthesia, until blood pressure de-

creases to the desired level, were tested. The therapeutic measures were tested after the viscera had been replaced and after determining the curve of the blood pressure.

The treatment of shock is described under four headings:

1. General measures. Heat, keeping the head down, etc. The value of the classical use of heat as well as the effect of cold in helping to produce the condition, was corroborated experimentally.

2. Special measures. Strapping the limbs, rebreathing, etc. Experimentally, rebreathing was not found to be of importance.

3. The use of drugs. Stimulants, vasoconstrictors. None of the drugs usually employed in the treatment of shock were found to be very effective.

4. The restoration of fluid volume. The best results in the treatment of experimental shock were obtained by the injection of fluid media. The data of the experiments justify the conclusion that none of the artificial solutions give such good results as the use of blood. The so-called colloidal solutions and their various modifications give better results than normal salt solution, but their potency is certainly not equal to blood or blood serum and occasionally they might be harmful.

BIBLIOGRAPHY

(1) Bayliss, W. M.: Methods of raising a low arterial pressure. Proc. Royal Soc., London, Series B., 1915–1917, lxxxix, 380.; Intravenous injection in wound shock, London, Longmans, 1918. 172 p.

(2) Bogart, L. J., Underhill F. P. and Mendel, L. B.: The regulation of the blood volume after injections of saline solutions. This Journal, 1916, xli, 189–218.

(3) Burnham, A. C.: The administration of glucose solutions as a prophylactic against post-perative shock. Amer. Journ. Med. Sci., 1915, cl, 431–436

(4) Cannon, W. B.: Shock and its control. This Journal, 1918, xlv, 544–545.

(5) Dale, H. H. and Laidlaw, P. P.: Surgical shock and some allied conditions. Brit. Med. Journ., 1917, i, 381–383.

(6) Dawson, P. M.: The changes in the heart rate and blood "pressures" resulting from severe haemorrhage and subsequent infusion of sodium bicarbonate. Journ. Exper. Med., 1905, vii, 1–31.

(7) Erlanger, J. and Gasser, H. S.: Hypertonic gum acacia and glucose in the treatment of secondary traumatic shock. Ann. Surg., 1919, lxix, 389–421.

(8) Erlanger, J. and Woodyatt, R. T.: Intravenous glucose injections in shock. Journ. Amer. Med. Assoc., 1917, lxix, 1410–1414.

(9) Fox, J. F.: The prevention and treatment of surgical shock. Amer. Journ. Surg., 1918, xxxii, 143–146.

(10) Gasser, H. S., Meek, W. J. and Erlanger, J.: The blood volume changes in shock and the modification of these by acacia. This Journal, 1918, xlv, 547–548.

(11) Gatch, W. D.: Nitrous-oxid-oxygen anesthesia by the method of rebreathing. With special reference to surgical shock. Journ. Amer. Med. Assoc., 1910, liv, 775–780; The use of rebreathing in the administration of anesthesia. Journ. Amer. Med. Assoc., 1911, lvii, 1593–1599.

(12) Gesell, R.: Observations on the volume flow of blood through the submaxillary gland. This Journal, 1918, xlv, 545–546.

(13) Guthrie, C. C.: The interpretation of the manifestations of shock. Penn. Med. Journ., 1918, xxii, 123–126.

(14) Henderson, Y. and Harvey, S. C.: Acapnia and shock. VIII. The venopressor mechanism. This Journal, 1918, xlvi, 533–553.

(15) Hogan, J. J.: The intravenous use of colloidal (gelatin) solutions in shock. Journ. Amer. Med. Assoc., 1915, lxiv, 721–726.

(16) Howell, W. H.: Observations upon the cause of shock, and the effect upon it of injections of solutions of sodium carbonate. Contrib Med. Research (Vaughn), Ann Arbor, Mich., 1903, 51–62.

(17) Mann, F C.: The peripheral origin of surgical shock. Johns Hopkins Hosp Bull., 1914, xxv, 205–212.

(18) Mann, F. C.: Shock and haemorrhage. An experimental study. Surg., Gynec. and Obst., 1915, xxi, 430–439.

(19) Mann, F. C.: Further study of experimental surgical shock Journ. Amer. Med. Assoc., 1918, lxxi, 1184–1186.

(20) Mann, F. C.: Studies on experimental surgical shock. This Journal, 1918, xlvii, 231–250.

(21) Meek, W. J. and Gasser, H. S.: The effects of injecting acacia. This Journal, 1918, xlv, 548–549.

(22) Porter, W. T.: Respiratory suction an aid in surgical shock. Boston Med. and Surg. Journ., 1917, clxxvi, 699.

(23) Seelig, M. G., Tierney, J. and Rodenbaugh, F.: An experimental study of sodium bicarbonate and other allied salts in shock. Amer. Journ. Med. Sci., 1913, cxlvi, 195–204.

EXPERIMENTAL STUDIES ON THE REGULATION OF BODY TEMPERATURE

III. The Effect of Increased Intracranial Pressure on Body Temperature

(Preliminary Communication)

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In a previous paper I (1) reported a series of "heat puncture" experiments on rabbits, the results of which indicated that in certain cases hyperthermia followed puncture of the brain, but since there was no correlation between the location of the lesion and the occurrence of the hyperthermia, the rise in temperature did not depend upon injury to the corpus striatum or other special "center." The existence of special "centers" in the brain for the regulation of body temperature was, therefore, not confirmed.

The hyperthermia which occurred in 22 per cent of the cases, however, remained unexplained, as also did the fatal symptoms accompanying a number of these and other cases of puncture; the symptoms noted in such cases suggested the possibility of a condition of increased intracranial pressure due perhaps to hemorrhage in the cranial vessels. This possibility was tested out by a series of experiments in which the intracranial pressure in rabbits was increased artificially. The symptoms produced by greatly increasing the pressure, 250 mm. or more of water, were identical with those observed in fatal puncture cases; increase in rate of respiration, slowing of heart beat, vasoconstriction, pupilo-dilatation, rise in body temperature followed by a fall before death. A moderate increase (150 to 200 mm. of water) produced less marked effects, the most apparent being an increase in the rate of respiration and vasoconstriction with a subsequent rise in body temperature. Practically every case with a definite increase in the pressure in the cranial cavity showed a rise in temperature sufficiently high to

be considered experimentally produced. The findings accord with the well-known symptoms in clinical cases of traumatic brain lesions.

The explanation of this influence of increased intracranial pressure on body temperature and also the fatal symptoms of higher pressure would appear to be that the pressure acts as a stimulus to the principal bulbar centers causing an increased rate of respiration by stimulation of the respiratory center, decrease in heart rate by stimulation of the cardio-inhibitory center, vasoconstriction by stimulation of the vasomotor center, and dilatation of the pupils by stimulation of the cervical sympathetic. In case of greatly increased and prolonged pressure these centers finally become paralyzed, their functioning ceases and death follows. Dixon and Halliburton (2) offer a similar explanation of the pressure symptoms observed by them in their experiments with dogs. On the other hand, with a moderate increase in intracranial pressure there is less evidence of bulbar stimulation, the most apparent excitation beng that of the vasomotor center. The vasoconstriction resulting from this stimulation causes more heat to be retained in the body and consequently a rise in the body temperature. In fatal cases the body temperature falls again just preceding death probably because of the paralysis of the vasomotor center.

These results support the view that temperature regulation is dependent upon physico-chemical factors without the intervention of hypothetical "heat centers."

The rise in body temperature and other symptoms attending increased intracranial pressure correspond so closely to those of "heat puncture" in which there is generally sufficient brain lesion to cause an increase in pressure in the brain cavity, and those of clinical brain lesions, that it seems possible to apply the same explanation to each of these cases. I think the rise in temperature which is reported by advocates of the "heat center" theory as due to "heat puncture" can be explained in a like manner, as can also the rise obtained in 22 per cent of my punctures and the fatal symptoms in a number of these and other cases.

The detailed results of this and further investigations will be published in a later paper.

BIBLIOGRAPHY

(1) Moore: This Journal, 1918, xlvi, 253.
(2) Dixon and Halliburton: Journ. Physiol., 1914, xlviii, 317.

STUDIES IN SECONDARY TRAUMATIC SHOCK

V. Restoration of the Plasma Volume and of the Alkali Reserve

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The method of estimating the blood volume employed in the preceding paper (1) brought to light the fact that the intravenous injection of a concentrated solution of gum acacia tends to prevent the concentration of the blood which otherwise practically invariably develops while shock is being induced. This observation suggested the present set of experiments which had as their first object a more detailed inquiry into the mechanism of this action of gum acacia, and ways of facilitating it.

SERIES I. OBSERVATIONS ON NORMAL ANIMALS

Procedure. Three experiments were performed on each of four normal dogs. The animals were anesthetized with morphine and ether. The first experiment on each dog consisted in the administration of a hypertonic crystalloid (18 per cent glucose, usually 5 cc. per kilo of body weight). After the animal had recovered from the effects of this injection, usually on the next day, a hypertonic colloid (5 to 6 cc. of a 30 or 25 per cent solution of gum acacia, respectively) was given. An interval of 3 days was then allowed to elapse. This was regarded as sufficiently long to permit of the disappearance of the gum acacia. Then in three of the instances the animal was given, first, the acacia and, immediately after, the glucose, both in the same concentration and amount as had been previously employed. To the fourth animal was given a 7 per cent solution of gum acacia. All injections were made as rapidly as possible. The hemoglobin percentage, determined as described in the preceding paper (1), served to indicate the effects these injections had upon the blood volume. The injections were made into the femoral vein and the samples of blood were taken

from some large artery. The arterial pressure was not followed in this series of experiments.

Results. When glucose is injected, the well-known fact is confirmed that the blood comes into osmotic equlibrium with the tissues within the first minute or two. The maximum theoretical dilution produced by the dose of glucose given may be calculated thus:—A 5.52 per cent solution of glucose is isotonic with blood

$$\left(\frac{6.88\ \text{(osmotic pressure of blood in atmospheres)} \times 18}{22.4\ \text{(osmotic pressure of 18 per cent glucose)}} = 5.52\right);$$

the 0.9 gram of glucose contained in each 5 cc. of the solution, the dose per kilo of animal, would therefore suffice to make $\frac{0.9}{0.0552}$, or 16.3 cc. of an isotonic solution. If we take 92 cc. as the quantity of blood in

TABLE 1

(1)	GLUCOSE			ACACIA			ACACIA AND GLUCOSE	
	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)
EXPERIMENT NUMBER	Dose per kilo	Theoretical increase in volume	Observed maximum volume increase	Dose per kilo	Theoretical increase in volume	Observed maximum volume increase	Dose per kilo	Observed maximum volume increase
		per cent	*per cent*		*per cent*	*per cent*		*per cent*
1	3 cc. 18%	10.6	7.0	5 cc. 30%	31.2	19.0	5 cc. 30% 3 cc. 18%	24.0
2	5 cc. 18%	17.7	6.0*	6 cc. 25%	31.2	10.0	6 cc. 25% 5 cc. 18%	18.0
3	5 cc. 18%	17.7	12.0	6 cc. 25%	31.2	11.0	6 cc. 25% 5 cc. 18%	35.0
4	5 cc. 18%	17.7	10.0	5 cc. 30%	31.2	12.0	18 cc. 7%	22.0

* Solution administered too slowly.

each kilo of animal the addition of 16.3 cc. per kilo will make a blood volume of 108.3 cc.; the blood volume is increased 17.7 per cent. Of the water required to effect this increase 5 cc. are furnished by the injection; the remaining 11.3 cc. must be taken from the tissues.

The results obtained are given in table 1, and two of the four sets of observations are plotted in figures 1 and 2. The table shows that in

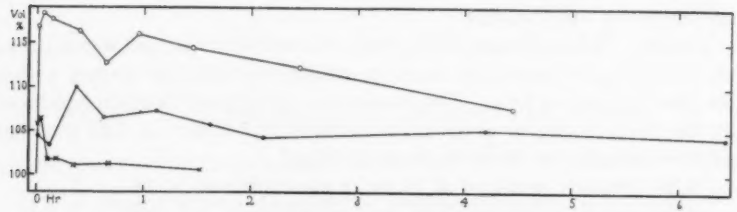

Fig. 1. Experiment 2, table 1. Blood volume, in per cent of the original, plotted against the time. Zero hour marks the completion of the injection of 5 cc. of 18 per cent glucose (—x—x—) on the 1st day, 6 cc. of 25 per cent gum acacia (— ●— ●—) on the 2nd day, and 6 cc. of 25 per cent gum followed immediately by 5 cc. of 18 per cent glucose (— ○— ○—) on the 4th day.

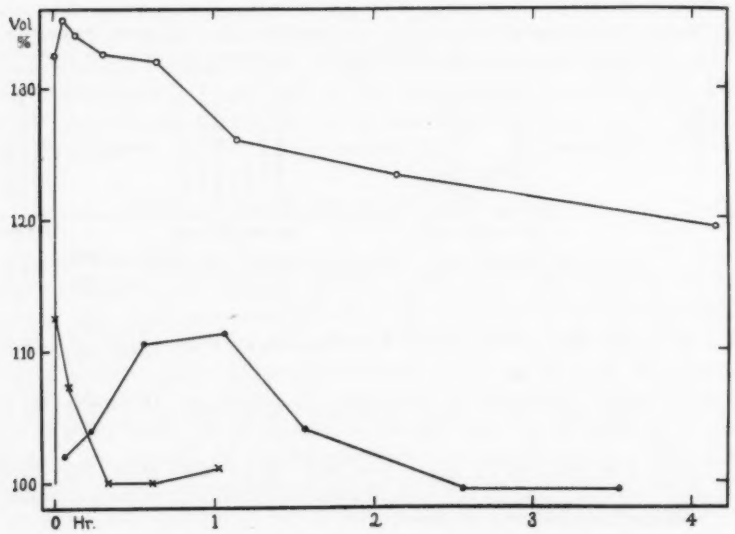

Fig. 2. Experiment 3, table 1. Blood volume in per cent of the original plotted against the time. Zero hour marks the completion of the injection of 5 cc. of 18 per cent glucose (—x—x—) on the 1st day, 6 cc. of 25 per cent gum acacia (— ●— ●—) on the 2nd day, and 6 cc. of 25 per cent gum followed immediately by 5 cc. of 18 per cent glucose (— ○— ○—) some days later.

each instance the observed maximum dilution after the injection of glucose (average = 8.7 per cent) falls far short of the theoretical maximum (17.7 per cent). This difference, presumably, is merely the result of the rapid loss of glucose from the circulation; it may be disappearing while the injection is proceeding, and also during the short interval elapsing between the termination of the injection and the taking of the first sample of blood. The largest discrepancy is seen in experiment 2, and in this instance it was recorded that the injection proceeded too slowly.

The maximum dilution was attained within ½ to 2 minutes. The blood then began to concentrate, rapidly at first, and then more slowly, and became normal within 5 to 45 minutes.

The results *following the injection of the acacia* solution differed quite decidedly from those obtained with the glucose. We present first our method of calculating the anticipated dilution of the blood as based upon the osmotic pressure of the acacia solution. According to data presented in the preceding paper (1) we can consider the osmotic pressures of 7 per cent acacia and blood serum 22 mm. and 16.4 mm. of mercury, respectively. On this basis 5 cc. of 30 per cent acacia would have to expand to 28.7 cc. to have the same osmotic pressure as the serum colloids. This would entail a 31.2 per cent expansion of the blood volume (normal 92 cc. per kilo).

The maximum dilutions observed experimentally (19, 10, 11 and 12 per cent) were in every instance much less than the theoretical 31.2 per cent. The dilution of 19 per cent was obtained in an experiment in which acacia was injected as soon as the dilution resulting from the glucose had disappeared; in the other experiments the acacia was injected one to three days after the sugar. This maximum dilution in every instance was attained very much more slowly (in 25 to 50 minutes) than when the glucose was injected (in ½ to 2 minutes). Of greater interest, however, is the slow return of the blood concentration toward the normal. In the four experiments the return to normal required more than 3 hours, more than 6 hours, 2½ hours and 3 hours respectively.

It is obvious that the colloid attracts water very slowly compared with the crystalloid glucose; therefore, at no time during the period of observation does the serum regain its normal osmotic properties. In spite of the increased colloid content of the plasma the maximum volume reached is not maintained. The plasma volume is reduced toward normal in spite of its increased colloidal osmotic pressure. This is

most probably to be attributed to filtration resulting from the increase in capillary pressure that would arise from the plethora and the increased viscosity. As acacia can maintain for some time a volume greater than normal it is to be expected that it would be even more effective when injected into an animal whose blood volume had been previously depleted, the volume-compensating mechanism not being then called into play. This is an example of the well-known pharmacological principle that it is easier to change a function toward normal than away from normal.

It follows from these observations that neither the concentrated solution of gum acacia nor the concentrated solution of glucose is perfectly satisfactory as a means of expanding the blood volume; the former attracts water slowly, though it holds it in the circulation for relatively long periods of time, whereas the latter attracts water with great rapidity, but the increased blood volume disappears with corresponding rapidity. To maintain an increased volume by the injection of glucose, the injection must be continuous.

It was conceived, therefore, that the shortcomings of each of these methods of expanding the blood volume might be corrected by combining the two; that if concentrated acacia were injected it would hold the water which a subsequent injection of concentrated glucose brought it. Experiment proved this to be true.

When glucose is injected after acacia we might expect, if the acacia holds all the water that the sugar can bring in, that the maximum theoretical blood volume increases of 10.6, 17.7 and 17.7 per cent respectively in the first three experiments would be added to the blood volumes resulting from the acacia injection. If we add the former percentages to the percentage increases obtained experimentally in the acacia control series (columns 3 and 7, table 1) we have theoretically possible expansions of 29.6, 27.7 and 28.7 per cent, that is, volumes within the theoretical holding power of the injected acacia. The corresponding expansions found were respectively 24, 18 and 35 per cent (column 9). Explanation of the variations from theory would demand a longer and more carefully controlled series. The data, at any rate, clearly support the original supposition, that glucose would bring in fluid which would be held by the acacia. The maximum dilution was reached very quickly; indeed, in two of the instances it was attained within 4 minutes or practically as quickly as when glucose was injected alone. In the other case the maximum was attained with the reading made 16 minutes after the injection, but 8 minutes had elapsed between this and the preceding reading.

After attaining the maximum, the blood volume at once begins to decline. In the two instances in which the maximum was quickly attained (experiments 2 and 3) this decline was slowly progressive as long as it was followed. At the time of the last readings made, the blood volume has always been higher following the injection of the two solutions than in the case of the injection of acacia alone. This is probably due in part to the greater initial increase in volume when the gum and the crystalloid are injected together, although it has been our experience in several instances that the increase in volume resulting from a rapid injection of very hypertonic colloid may be rapidly disposed of. The reason for this we have not analyzed.

The slow return of the blood volume to normal is seen also after the injection of more dilute (7 per cent) acacia. In the third part of experiment 4, 18 cc. per kilo of such a solution were injected, a volume sufficient to increase the blood volume (normal 92 cc. per kilo) 19.6 per cent. The actual dilution observed was about 21 per cent directly after the injection. The blood volume began to decline at once, but so slowly that 5 hours later it was still 111 per cent of the original. The 7 per cent acacia was chosen in this experiment on the assumption that we were working with an isotonic colloid. According to our present calculations, however, even the 7 per cent would be appreciably hypertonic.

SERIES II. OBSERVATIONS ON ANIMALS IN SHOCK

1. The effect of injection of solutions of colloids and crystalloids on the circulation

The main conclusion to be drawn from the experiments reported in part 1 of this paper is that in normal animals a hypertonic colloid will hold in the circulation not only the water that it itself slowly attracts, but also the water brought to it rapidly by a hypertonic crystalloid, so that the combined injection of the two results in a rapid and well-sustained expansion of the blood volume. The colloid employed in those experiments was gum acacia; the crystalloid, glucose. In view of the acidosis demonstrated to be present in the types of shock with which we were working it was deemed advisable in the tests carried out on animals in shock to employ as the crystalloid either Na_2CO_3 or $NaHCO_3$, instead of glucose. It may be assumed that the principles involved, as regards changes in the blood volume, are not altered by this change in the crystalloid employed.

Methods. The animals were first traumatized by holding the arterial pressure down to 40 mm. Hg. for a period of about 2 to 2¼ hours by partly occluding the inferior vena cava (2), (3). This damage in our experience in time usually brings on a shock-like failure of the circulation. The blood volume was followed, as previously, by estimating the hemoglobin. In addition, the pressure in the carotid artery was followed by a method that entailed practically no loss of blood or mixture of the anticoagulant salt with the blood in the animal. In a few experiments the CO_2 capacity of the blood was followed by the Van Slyke method.

In most of the experiments the solution injected was 25 per cent sodium acacia in 4 or 5 per cent Na_2CO_3 or $NaHCO_3$, the amount, 4 or 5 cc. of the solution per kilo of body weight, though many other combinations, including the first Bayliss solution (4), were used. As a 1.37 per cent solution of Na_2CO_3 is isosmotic to blood, each cubic centimeter of a 5 per cent solution injected would have to expand to 3.65 cc. before becoming isosmotic to plasma. Our 5 per cent solution of $NaHCO_3$ lowered the freezing point of water 1.879°C. One cubic centimeter of this would, therefore, expand to about 3.35 cc. Five per cent solutions of these substances are, therefore, comparable with respect to their osmotic pressures to the 18 per cent glucose in the preceding experiments. Owing to the inadvisability of rapidly injecting concentrated acacia solutions into animals in shock, the injection in these experiments was prolonged usually to 20 or more minutes.

Blood volume. The changes in the blood volume that occurred are collected in table 2. In every case the increase in volume after the injection of 25 per cent gum in combination with 4 or 5 per cent carbonate is at least as great as that calculated to be necessary for all the crystalloid injected to become isosmotic. And in no instance is the increase in blood volume, determined shortly after the injection, any greater than the volume of water that the injected gum acacia is theoretically capable of holding. But in experiments 22, 25 and 30 the blood volume continues to increase so that the ultimate dilution is greater than the theoretical holding power of the acacia, though within the limit of error of the methods, in the case of experiment 30. The explanation of this result is not obvious. There is the possibility that the improvement in the circulation permits the organism to participate in increasing the blood volume.

A glance at the table will show that other solutions of gum acacia, such as simple 7 per cent gum acacia and 6 per cent gum acacia in

TABLE 2

EXPERIMENT NUMBER	TREATMENT	SHOCK VOLUME	BLOOD VOLUME CHANGES FROM INJECTION						ARTERIAL PRESSURE			RESULT
			Within 3 minutes	Maximum			Last observation					
				Volume	Time after injection	Increase	Volume	Time after injection	Initial	Before treatment	Immediately after treatment	
		per cent of normal	*per cent of normal*	*per cent of normal*	*minutes*	*per cent*	*per cent of normal*	*minutes*	*mm. Hg.*	*mm. Hg.*	*mm. Hg.*	
22	25% acacia in 5% Na_2CO_3, 5 cc. per kilo	90.2	115.8	131.0	67	45.0	125.2	120	105	105	130	Died
25	25% acacia in 5% Na_2CO_3, 5 cc. per kilo	84.0?	115.0	127.2	78	51.4?	127.2	78	140	45	70	Died
24	25% acacia in 5% Na_2CO_3, 4 cc. per kilo	87.6	104.2	104.2	2	19.0	98.2	121	120	95	130	Lived
30	25% acacia in 5% Na_2CO_3, 4 cc. per kilo	85.6	109.0	112.6	100	31.6	112.6	100	130	80	84	Died
34	25% acacia in 4% $NaHCO_3$, 4 cc. per kilo	80.0	94.0	94.0		17.4	94.0	62	120	95	107	Died
43	25% acacia in 4% $NaHCO_3$, 5 cc. per kilo	75.9	91.9	91.9		21.0	91.9	6		100	115	Lived
41	25% acacia in 4% $NaHCO_3$, 4 cc. per kilo	80.0	100.4				100.4	24	105	92	100	Died
18a	7% acacia in 2.44% Na_2CO_3, 24 cc. per kilo	88.8	122.4	122.4	3	37.8	111.0	128	105	90	110	Died
28	6% acacia in 2% $NaHCO_3$, 12 cc. per kilo	85.7	111.7	114.3	13	33.4	114.0	13	120	58	68	Died
18	6% acacia in 2% $NaHCO_3$, 18 cc. per kilo	86.8	108.2	113.2	109.2	30.4			110	98	115	Died
17	7% acacia, 18 cc. per kilo	81.6	94.8	95.8	32	17	92.2	80	180	187	80–120	Died

2 per cent $NaHCO_3$ (4), when osmotic properties as well as volume of fluid injected are taken into account, are quite as efficacious in restoring and maintaining the blood volume as are the stronger solutions. While there is often some recession from the maximum dilution this is never large, and in five of seven animals in which the concentration was followed for over 1 hour after the injection, the volume at the last reading is as large as that obtaining within the first 3 minutes after the injection.

When these experiments were performed we believed that 7 per cent acacia was isosmotic with the colloids of the blood and that its effect on the blood volume was merely additive. On the basis of data presented in the preceding paper (1), we now consider the colloidal osmotic pressure of serum to be 16.4 mm. of mercury and of 7 per cent acacia 22 mm. We have recently dialyzed a 7 per cent solution of acacia in the same Locke's solution used in making the above determinations, against serum in a Moore and Roaf osmometer, arrangement being made for intermittent renewal of the serum in the lower chamber. The pressure developed was 6.6 mm. of mercury. The pressure on the acacia was released after the first determination and the experiment then continued with the salts in the acacia solution modified by the interchange of salts with the serum during the first determination. The pressure developed in the second determination was again 6.6 mm. This pressure is to be compared with the difference of 5.6 mm. between the separate direct determinations.

After the injection of 18 cc. per kilo of 7 per cent acacia the osmotic pressure of the plasma colloids is calculated to be 8.5 per cent above normal. After the injection of 5 cc. per kilo of a solution containing 18 per cent glucose and 25 per cent acacia the colloids would be 10.7 per cent hypertonic after enough fluid had been brought in from the tissues to render the crystalloid isotonic, assuming that all the glucose stayed in the vessels till diluted. It therefore follows that after the injection either of the 7 per cent acacia solution or of the very hypertonic gum and glucose solutions in the above quantities the blood plasma is left with a colloidal osmotic pressure somewhat above normal.

In the solution containing 6 per cent acacia and 2 per cent $NaHCO_3$ both the colloid and crystalloid are slightly hypertonic, even after making allowance for the loss of carbonate in the precipitation of $CaCO_3$. Table 2 shows that the blood dilution resulting from the injection of the 6 per cent gum acacia solution in 2 per cent $NaHCO_3$ was greater

than could be accounted for simply by the addition of the volume of water actually injected.

Blood pressure. Practically invariably the arterial pressure is raised by the injection of any of these solutions containing gum and crystalloid (see table 2) even when the injection of the crystalloid alone is without effect (see *a*, fig. 6). The effect of the injection upon the blood pressure, however, seems to be merely an incident in the course taken by the pressure as determined by the reaction of the animal to the clamping of the cava. If at the time the gum solution is injected the pressure is rising, the injection facilitates the rise somewhat; but if the pressure is falling, the solution while usually causing the pressure to rise for a time, fails to maintain the pressure. Sooner or later the pressure begins to fall again, and the animal invariably dies. This fall in pressure may occur even while the blood volume apparently is increasing.

2. Alkali reserve in experimental shock

Before presenting the effect on the alkali reserve of the acacia-carbonate mixtures, the effect of the shock-producing procedures which we have employed, upon the carbonate content of the plasma should be recorded. This study was made because at the time these experiments were performed considerable importance was being attached to the reduction in reserve alkali which had been found to obtain in clinical shock (5).

In a number of animals we have followed with the Van Slyke apparatus the changes in the CO_2 capacity of the plasma of arterial blood drawn without loss of CO_2. This is regarded by Van Slyke and Cullen (6) as the ideal method of estimating the alkali reserve. Most of the determinations were made in duplicate.

The data are collected in figure 3 in which the volume per cent of CO_2 is plotted against the mean arterial pressure. The first blood sample in each case was drawn after anesthetizing the animal with morphine followed by ether and after making all of the operations preparatory to starting the procedures by which shock-like failure of the circulation was to be induced. It is seen that these readings ranged between 50.5 and 26.1, and usually were below 40.0.

In order to avoid any complications that might arise as a result of the withdrawal of blood, it was necessary to reduce the number of readings of the CO_2 capacity to a minimum. As a rule, therefore, only one other reading was made and that at a time when we felt as-

sured that a shock-like failure of the circulation had become established. The readings made at that time, with but one exception (no. 2), show a markedly reduced CO_2 capacity. And, with but this one exception, the inclination of the line joining the initial with the shock reading in almost every instance is remarkably constant; these lines are all nearly parallel to each other.

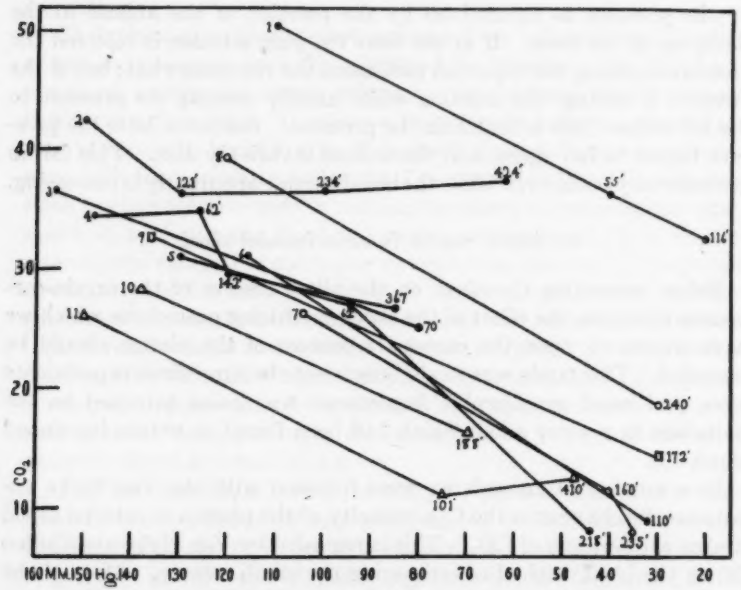

Fig. 3. Showing the relation of CO_2 capacity of arterial blood (ordinates) to arterial pressure (abscissae). The first reading in each case is made before starting to induce shock; the subsequent readings later by the time (in minutes) indicated in each case. Shock by intestinal exposure, —●—; shock by caval occlusion, —○—; shock by aortic occlusion, —□—; shock by adrenalin injection —△—.

On the other hand the gradient of the curves or of the parts of the curves joining readings made before the fall in pressure in general had exceeded 50 per cent of the original value is not quite so steep. This possibly may mean that the tissue alterations that lead to the reduction in alkali reserve do not progress rapidly until the condition that brings them about has reached a relatively advanced stage of develop-

ment. This inference is borne out in the main by the curves of the relation obtaining between the CO_2 capacity and the volume flow of blood published by Gesell (7). His curves as a rule show that the flow of blood through the salivary gland may be below 50 per cent of the original for some time before the CO_2 capacity begins to fall. It would seem, therefore, that the reduced CO_2 capacity is the result and not the cause of the fall in arterial pressure.

The gradient of the line joining the initial with the final readings as may be seen in figure 3 is largely independent of the time. It can also

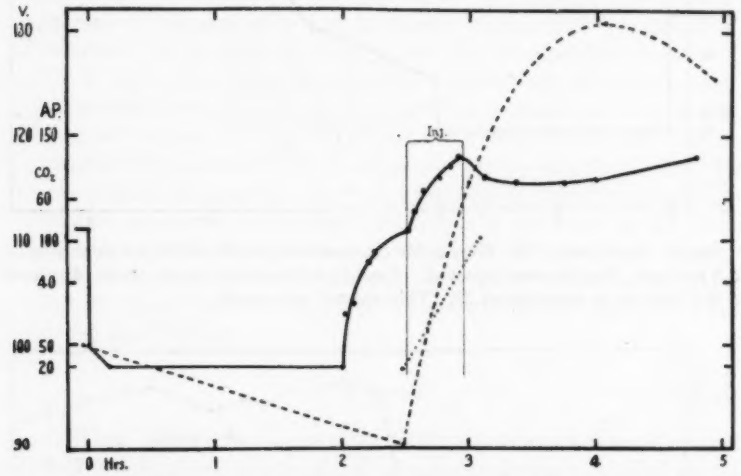

Fig. 4. Experiment 22. Arterial pressure, —●—●—; blood volume, - - ●- - ●- -, per cent of normal, determined by the reciprocal of the Hb. content; CO_2 capacity, ··●···●·· The figure shows the acceleration of the rise of pressure, that follows release of the cava, by the injection of 5 cc. per kilo of a solution consisting of 25 per cent sodium acacia in 5 per cent Na_2CO_3, and the subsequent maintenance of a good pressure. It shows, also, a tremendous increase in the blood volume resulting from the injection. The blood volume is still high at the close of the experiment, more than 2 hours after completing the injection. It also shows the rise in the CO_2 capacity. This animal died before twenty-three hours had elapsed.

be seen that though the lines tend to parallel each other they may be on widely separated levels, so much so that the final readings in a case of advanced shock may be within, or almost within, the range of the normal readings of other cases. While there is no clear direct relationship

between the CO_2 capacity and the arterial pressure, it nevertheless is obvious that a reduction in the carbonate content of the plasma from arterial blood is a constant accompaniment of shock as we have induced it.

The alkali reserve after injections of solutions containing acacia and carbonate. The carbonate content of the arterial blood was followed in two of the cases that received 5 per cent Na_2CO_3 with the acacia.

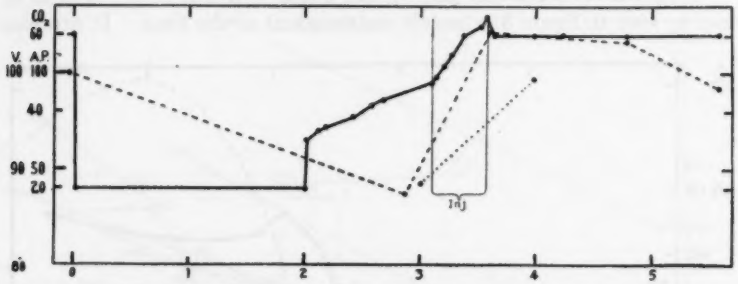

Fig. 5. Experiment 24. Four cubic centimeters per kilo of 25 per cent acacia in 5 per cent Na_2CO_3 were injected. Essentially the same results were obtained in this case as in experiment 22. This animal recovered.

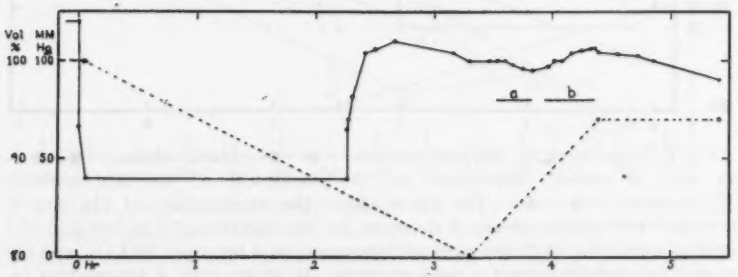

Fig. 6. Experiment 34. In this case the pressure began to fall when something less than 54 minutes had elapsed after removing the clamp from the cava. At a time when the pressure was stationary at 100 mm. of Hg. the animal (at *a*) was given 4 cc. per kilo of a 4 per cent solution of $NaHCO_3$. The pressure fell somewhat while this was being given. A solution consisting of 25 per cent sodium acacia in 4 per cent $NaHCO_3$, 4 cc. per kilo, was then given (at *b*). Under the influence of this injection the pressure rose and the blood volume increased, but neither was quite restored to normal. Almost immediately after terminating the injection the blood pressure began to fall again despite any measureable decrease in blood volume. This animal died.

One received 5 cc. per kilo of body weight, the other 4 cc. In the first case (fig. 4) the volume of combined CO_2 was raised from the shock level of 19.8 per cent to 50.6 per cent; and in the second case (fig. 5) from 20.6 per cent to 48.6 per cent. The values attained in each case, therefore, were quite normal for dogs under ordinary conditions of experimentation.

The ultimate results of the injection of a solution containing hypertonic acacia and carbonate. A general discussion of the effect of the injections on the blood pressure, the blood volume and the alkali reserve has been given. There remains to be considered the fate of the individual animals. In figures 4, 5 and 6 typical experiments are graphically presented. It is obvious from these and similar experiments (see table 2) that an intravenous injection which raises the blood pressure, and maintains the rise for the period of the experiment, which restores the blood volume and the alkali reserve in an animal that has been damaged to an extent that in time usually results in a shock-like failure of the circulation, may not prevent a fatal issue. One cannot help but conclude, therefore, that these alterations in the state of the animal do not constitute the primary cause of death from shock; in other words, they are merely symptoms of some more fundamental process, which determines the condition of the animal but to which the former may be contributary.

SUMMARY

1. When glucose in 18 per cent solution is injected into the circulation of a normal animal the blood comes into osmotic equilibrium with the tissues within the first minute or two, the average maximum dilution amounts to but half of the theoretical maximum and the blood regains its normal concentration within 5 to 45 minutes.

2. When gum acacia in a concentrated solution is injected the average maximum dilution of 41.7 per cent of the theoretical maximum is attained within 25 to 50 minutes; the decline of the blood volume to normal requires $2\frac{1}{2}$ to 6 or more hours.

3. When the concentrated acacia is immediately followed by the glucose the maximum dilution is quickly attained and is much greater than that resulting from the injection of either of the two substances alone. The dilution is well maintained.

4. Comparable results are obtained in animals in shock when a strong solution of gum acacia is followed by a solution of Na_2CO_3 that is isosmotic to 18 per cent glucose.

5. With such a combination of solutions given in appropriate amounts the blood volume, the blood pressure and the reserve alkali of animals in shock often can be brought to normal and held there for the usual duration of an experiment. Yet such animals, as well as shocked animals treated with other combinations of gum acacia and carbonate or bicarbonate, often died within 24 hours.

BIBLIOGRAPHY

(1) GASSER AND ERLANGER: This Journal, 1919, l, 31.
(2) JANEWAY AND JACKSON: Proc. Soc. Exper. Biol. and Med., 1915, xii, 193.
(3) ERLANGER AND GASSER: This Journal, 1919, xlix, 151.
(4) BAYLISS: Private Rept. of the English Committee on Shock, November 29, 1917.
(5) CANNON: Journ. Amer. Med. Assoc., 1918, lxx, 531.
(6) VAN SLYKE AND CULLEN: Journ. Biol. Chem., 1917, xxx, 366.
(7) GESELL: This Journal 1919, xlvii, 468.

STUDIES IN SECONDARY TRAUMATIC SHOCK

VI. Statistical Study of the Treatment of Measured Trauma with Solutions of Gum Acacia and Crystalloids

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INTRODUCTION

With the growth of our personal experience with experimental shock we came more and more to feel that a solution consisting of hypertonic gum acacia and of a hypertonic crystalloid would prove helpful in counteracting the disturbances which seemed to be responsible for the failure of the circulation that constitutes the main feature of that state.

The growth of this idea may be briefly outlined. Our first experiments had as their object a study of the hemodynamics of shock. They convinced us that a giving way of the vasomotor center or of the heart is to be regarded, speaking broadly, only as a relatively late consequence of a low arterial pressure, and not as its cause. In the early stages of shock as induced by exposure of the intestines (1), by partial occlusion of the aorta or of the vena cava (2), or by the administration of adrenalin (3), the only constant circulatory fault seemed to be a reduction in blood volume, both actual and effective (4). We found in all types of shock, among other less constant lesions, marked dilatation of the capillaries and venules of the villi of the intestines.

Since our methods of inducing shock all seemed to depend upon the effects of slowing the blood stream, since furthermore the same vascular changes can be seen to develop during partial occlusion of an artery (5), and since the shock that follows the administration of adrenalin seems to be referable to the constriction of the arterioles it produces by local action, we ventured to suggest (6) that in man the

[1] With the assistance in some of the experiments of Paul C. Hodges.

prime factor, also, might be a strong vasoconstriction compensating hemorrhage and wound weeping, and aggravated by exposure and pain. The changes thus produced in the capillaries and venules by the slowing of the circulation would account for the concentration of the blood found in experimental shock by attributing it to transudation of plasma and they account for the reduction in effective blood volume through stagnation in the dilated vessels.

Although the disappearance of plasma from the circulating blood is of constant occurrence in shock experimentally produced, there is evidence that while plasma is disappearing, fluids are being added to the blood (4). The latter process may be regarded as an effort on the part of the organism to combat the diminution in blood volume. Evidence has been obtained indicating that the ability of the organism to thus make good the loss in blood volume decreases as shock develops.

It was while we were studying the blood volume in shock that we happened upon the observation that the concentration of the blood that develops during shock induction does not occur, or at least is not so marked, when shock is induced after the administration of a dose of hypertonic gum acacia. This result we were able to show is due in part, at least, to the osmotic pressure the hypertonic gum exerts (7). It was found, however, that hypertonic gum solutions attract tissue fluids into the circulation very slowly. Hypertonic crystalloids injected intravenously, as is well known, attract water quickly. When the two, the hypertonic gum and the hypertonic crystalloid, are injected to all intents and purposes, simultaneously, the gum holds the water the crystalloid quickly brings into the circulation. Presumably, the blood volume is thus increased by a process that resembles the one the organism itself employs in combatting a reduction in blood volume. Inasmuch as the reserve alkali was found to be reduced in the types of shock we were studying (7), it was felt that sodium bicarbonate or sodium carbonate might be made to act at one and the same time as the hypertonic crystalloid and as a supply of base. We therefore studied the action on animals in shock of hypertonic gum acacia and hypertonic bicarbonate, as well as other combinations of gum and crystalloid and found (7) that while all equivalent combinations acted on the arterial pressure, the blood volume and the alkali reserve approximately alike and about as had been anticipated, many of the animals, nevertheless, died.

It thus became obvious that in order to ascertain whether these or other similar solutions that have been proposed or may be proposed

for the intravenous treatment of shock are of value, it would be necessary to study their action on a standard condition, one whose course, if left to itself, could be predicted with a reasonable degree of certainty and to use as the criterion of their efficacy not their effect on the blood pressure, or on the blood volume, or on the alkali reserve, but actual recovery from the state of shock. In the present state of uncertainty of our knowledge with regard to the nature of shock, no other criterion can be convincing. It was felt that the conditions for such a study would be supplied best by exposing an animal to a form of trauma which in time brings on a shock-like failure of the circulation, and to so grade that injury that it would just cause death if things were allowed to take their course. To state this conception in another way, the interpretation of therapeutic tests would be entirely free of ambiguity if they could be made on animals on which a minimal fatal amount of injury had been inflicted, and if recovery from the immediate effects of the damage were taken as the criterion of efficacy.

Of the various shock-producing procedures with which we were familiar only two seemed likely to yield to such standardization, namely, partial temporary occlusion of the aorta or of the inferior vena cava. Exposure of the abdominal viscera has been so used by Mann (8), but we doubt if such damage can be uniformly applied. Other forms of mechanically produced tissue damage are very uncertain in their effects or cannot be inflicted aseptically (9); massive doses of adrenalin do not always cause death through shock; and it is still very questionable whether acapnia (10) or fat embolism (11) bring on shock properly so-called, either clinically or experimentally.

THE STANDARD DAMAGE

After a few trials we decided to use as the damage partial occlusion of the vena cava (12). The exact procedure finally adopted as the result of a number of preliminary trials was as follows: Under morphine and ether anesthesia, and under strict asepsis, a clamp, adjustable by means of a finely threaded screw, is placed on the inferior vena cava of the dog between the diaphragm and the liver, and the vein is so compressed as to attempt to hold the arterial pressure at 40 mm. Hg. for a period of 2 hours 15 minutes. The ether is discontinued after applying the clamp, and owing to the apathetic condition of the animal, it need not be administered again.

As a rule the arterial pressure can thus be brought down to 40 mm. Hg. at once and held there. But sometimes the arterial pressure at

first does not fall to 40. In such instances, however, the pressure as a rule tends to fall, and usually reaches 40 mm. Hg. in the course of a few minutes, when the clamp can be opened and adjusted so as to hold the pressure at 40. Occasionally, however, the pressure falls quite slowly and in a few instances it has failed to reach the level of 40 mm. Hg., even by the end of the 2 hour and 15 minute clamping period. We have in several of the latter instances determined the position of the clamp by post-mortem examination and invariably have found that it was occluding the cava. Nevertheless, recognizing the danger of including in the series cases in which the position of the clamp may have been faulty, we have once and for all excluded all instances in which the pressure by the end of the period had failed to fall to within a millimeter or two of 40. It may be added that the pressure is just as apt to fail to fall when the initial arterial pressure is high as when it is low.

Experience has indicated that the cases in which difficulty is experienced in lowering the pressure are rather more apt to recover from the effects of the caval occlusion than cases in which the pressure can be brought down to 40 mm. Hg. at once. Indeed, in one or two instances the former, after removal of the clamp, have recovered without exhibiting any of the evidences of shock. The more favorable course of such cases seems capable of two explanations: either *a*, the high pressure protects the heart and medullary centers from the damage we have reason to believe (2) is done by the long-continued low pressure of the clamping period; or *b*, the high pressure of the clamping period is the result of an unusually good collateral circulation which protects the posterior parts of the body, as well as the anterior, from the effects of the occlusion. But whatever the explanation, the fact remains that failure of the pressure to fall seems to favor recovery. Some rule must, therefore, be made that will take this fact into account. We feel that recovery would be favored but little, if any, where the period during which the clamp is tight and the pressure above 40 mm. Hg. does not exceed 30 minutes. This arbitrary decision is applied in the statistical treatment of the experiments; it will be seen that the exclusion of such of these cases as recover does not materially alter the results.

When the clamp is removed the arterial pressure rises with a bound, sinks again almost at once, usually to somewhere between 50 to 75 mm. Hg., and then mounts slowly. Experience has shown that the animal will die if now the pressure begins to fall consistently, even

though slowly, before 2 hours have elapsed.[2] This outcome we have not succeeded in preventing by any of the forms of treatment we have employed. We therefore exclude from the series those cases in which the pressure begins to fall consistently before 2 hours have elapsed, and treatment is not begun until this two hour period of observation has passed.

One case (no. 215, table 7) has not been accepted although, strictly speaking, it is not excluded by this rule. The pressure in this case did not fall during, though it began to fall immediately after, the conclusion of the observation period and before treatment was begun. This has not occurred in any other case. Furthermore, the pressure at its highest did not get above 64 mm. Hg., a level which is 6 mm. below the lowest maximum observed in the whole series of 168 cases.

If the animal does not die within 48 hours, timed from the beginning of the clamping period, as a consequence of the damage done through clamping the cava, very marked improvement almost invariably is apparent particularly as regards the state of apathy.

In eight instances, however, animals that had shown this improvement were found dead unexpectedly, seven at the close, the eighth at the beginning, of the third day. Such animals, obviously, had recovered from the immediate effects of the injury, but died of other causes. They are, therefore, included in the category of those that recovered from shock. Our reasons for thus treating these cases will be discussed later.

A number of animals have become unavailable on account of certain accidents. Thus, animals that have died within 2 days and in which, at autopsy, extensive abdominal hemorrhage was found, must be discarded, for here the animal has had to contend not alone with the

[2] There has been but one exception to this rule, and that exception was not a clean-cut one. In the case of dog 208 (see table 3) after removing the clamp the pressure rose from the minimum of 55 mm. Hg. to a maximum of 97 mm. Hg. in the course of 27 minutes, but during the next 30 minutes it fell gradually to 80. Toward the close of the 2-hour observation period it began to rise, reaching 90 mm. Hg. 2 hours 10 minutes after removing the clamp. The animal's pressure, followed 2 hours longer, continued to rise, eventually reaching 95 mm. Hg. This animal recovered. It can scarcely be said that in this case the pressure began to fall *consistently* before the close of the 2-hour period; or as a matter of fact it eventually began to rise. It should be added that this was also a *cardiac* case (see below) and in the effort to carry the animal through the clamping period of 2 hours 17 minutes, it twice was necessary to partially open the clamp, once for a period of 8 minutes and once for a period of 2 minutes.

effects of the temporary anemia of the clamping period, but also with the effects of the hemorrhage. In most instances this hemorrhage was unavoidable and usually due to tearing of the liver through tension exerted by the clamp on an unusually broad suspensory ligament.

Cases exhibiting slight abdominal hemorrhage at autopsy have been included in the series. The decision as to whether a case that is on the border line is to be included in, or excluded from, the series must of necessity be arbitrary. We have therefore presented the results obtained after excluding every case showing abdominal hemorrhage of any degree whatever. It will be seen that this way of viewing the data affects them quite distinctly. Abdominal hemorrhage, it might be added, has been no more frequent in treated than in untreated animals. This fact is mentioned in order to allay any suspicion that the hemorrhage might be a consequence of treatment. We add, also, that we have observed no evidence of a tendency to bleed other than that attributable to the rise in pressure produced by the injection.

The control series, that is, the series of animals used for the purpose of determining the mortality rate from the standard damage, includes 23 "acceptable" cases. The acceptable group, to repeat, is made up by excluding cases in which the arterial pressure began to fall before the 2 hours had elapsed after removing the clamp, or in which at autopsy considerable abdominal hemorrhage was found. It is seen (table 1) that 52 per cent of the animals recovered. This method of doing damage, therefore, does not meet one of the conditions required of the ideal test; the damage inflicted does not just kill all the animals. We were unable, however, to come any closer to the ideal. When the clamping period was prolonged beyond 2 hours 15 minutes or when the arterial pressure was held below 40 mm. Hg., so as to get every untreated animal to die subsequently, treatment was absolutely without avail. The method adopted of preparing animals for treatment is not, therefore, as satisfactory as we had hoped to evolve, but though the method is not ideal, we felt convinced that by using a sufficiently large number of animals and by basing conclusions only on decided results, it would suffice our purposes. On the other hand the method has the advantage of indicating, through variations in the death rate, deleterious action as well as beneficial action.

Analysis of the control series, as well as of the pre-treatment stage of the several treated series, indicated the possibility of using in the interpretation of the results of treatment other data than those furnished by the number of deaths. Thus, it is seen that the span of

life in the fatal cases varies. It is obvious that if any particular form of treatment were injurious, its deleterious action might manifest itself not alone in an increase in the number of deaths but also in a shortening of the duration of the anti-mortal stage. As there were long periods during the night when the animals were not under observation, and as most of the animals, especially those dying within

TABLE 1

Summary of mortality statistics

TREATMENT	TOTAL NUMBER OF ANIMALS	ACCEPTABLE CASES	DEATHS WITHIN 48 HOURS		AFTER EXCLUDING SLIGHT HEMORRHAGE				DEATHS WITHIN 24 HOURS		EXCLUDING FATAL CARDIAC CASES			EXCLUDING CASES 30′ + ABOVE 40 MM. Hg. THAT LIVED		
							Deaths within 48 hours					Deaths within 48 hours			Deaths within 48 hours	
			Number	Per cent	Number	Left	Number	Per cent	Total	After excluding slight hemorrhage	Left	Number	Per cent	Left	Number	Per cent
1	*2*	*3*	*4*	*5*	*6*	*7*	*8*	*9*	*10*	*11*	*12*	*13*	*14*	*15*	*16*	*17*
Controls	40	23	11	48	4	19	7	36.9	4	1	22	10	45	18	11	61.2
6 in 2 per cent gum-bicarbonate	27	20	9	45	3	17	6	35.3	2	2	19	8	42	16 or 17	9	56.3 or 53.0
25 and 5 per cent gum-bicarbonate	27	16	9	56	2	14	7	50.0	8	6	15	8	53	13	9	69.3
25 and 18 per cent gum-glucose	43	20	9	45	4	16	5	31.3	1	0?	18	7	39	19	9	47.4
25 in 18 per cent gum-glucose	33	21	5	24	4*	18 or 17	2 or 1	11.0 or 5.9	3*	0	20	4	20	16	5	31.3

* One of these cases had no abdominal hemorrhage, but the pleura contained a large amount of bloody exudate and the liver was the seat of a chronic process.

24 hours, succumbed during the night, we find it impossible in most instances to give the duration of life in hours. It is, however, possible to accurately divide the fatal cases into those that die within 24 hours and those that die within 48 hours, and this we have done.

The arterial pressure also furnishes information of value in the analysis of the results obtained. Tables 2 and 3 show that in the control series the average initial arterial pressure is very much higher

in the case of the animals that died (109.8 mm. Hg.) than in the case of those that lived (89.4 mm. Hg.). Excepting one group, this is true also of the groups of animals that were subsequently treated. This observation can mean but one thing, namely, that a high initial pressure prejudices the animal's chances of recovery from the effects of holding its pressure down to 40 mm. Hg. for a fixed pericd. To what this may be due is a question we need not attempt to answer here.

TABLE 2

Summary of average pressures

			DIED					RECOVERED OF SHOCK				
	Initial pressure	Pressure after 2 hours	Number of cases	Initial pressure	Pressure after 2 hours	Pressure before treatment	Pressure last reading	Number of cases	Initial pressure	Pressure after 2 hours	Pressure before treatment	Pressure last reading
	1	*2*	*3*	*4*	*5*	*6*	*7*	*8*	*9*	*10*	*11*	*12*
Controls.....	99.6	102.9	11	109.8	102.4	105.0	105.0	12	89.4	103.4	109.2	109.2
6 in 2 per cent gum-bicarbonate.....	100.9	101.7	9	107.9	101.9	102.6	111.1	11	93.9	101.5	103.7	113.4
25 and 5 per cent gum-bicarbonate....	94.2	103.2	9	98.8	98.3	99.8	113.0	7	89.7	108.1	109.3	118.0
25 and 18 per cent gum-glucose.....	105.0	105.0	9	106.6	97.5	99.6	115.1	11	103.4	112.6	115.1	131.2
25 in 18 per cent gum-glucose.....	103.1	103.1	5	102.0	100.2	103.0	118.0	16	104.2	106.0	106.8	117.6
Averages.....	100.6	103.2		105.0	100.1	101.2	114.3*		96.1	106.3	108.7	120.0*

* Controls not included.

The important point is that on the average the animal with the high initial arterial pressure evidently is handicapped.

On the other hand it is seen, and for this purpose again all of the groups are available, that by the end of the 2-hour period of observation the arterial pressure of the cases that live, on the average, reaches a level considerably above the initial, whereas the average arterial pressure of the cases that die usually falls short of reaching the initial level. Presumably, therefore, speaking generally, a high post-decompression pressure favors recovery from the effects of clamping.

TABLE 3

Control series

DIED							RECOVERED OF SHOCK					
Serial number	Initial pressure	Pressure end of observation period	Last reading: Time after observation period	Last reading: Pressure	Slight abdominal hemorrhage	Remarks	Serial number	Initial pressure	Pressure end of observation period	Last reading: Time after observation period	Last reading: Pressure	Remarks
	mm.	*mm.*	*min.*	*mm.*				*mm.*	*mm.*	*min.*	*mm.*	
101	125.0	120.0	21	125	–		93	105.0	104.0	20	108.0	
96	120.0	111.0	31	112	–	8′ above 40	100	102.0	122.0	59	128.0	86′ above 40
98	120.0	90.0	25	93	–	7′ above 40	35	95.0	100.0	0	100.0	Blood pressure at 29 hours; 105 mm. Hg; animal seemed well; killed
103	118.0	108.0	15	112	+	Died within 24 hours	120	95.0	100.0	0	100.0	13′ above 40
92	115.0	98.0	12	99	–		208	95.0	88.0	95	95.0	4′ above 40. Cardiac; clamped 2 hours 17′, partly open 10′
111	110.0	100.0	12	103	–		102	90.0	100.0	23	110.0	
62	110.0	84.0	33	90	–	Cardiac; clamped 2 hours 16′, partly open ?′	115	90.0	100.0	24	112.0	
117	102.0	95.0	0	95	+		207	85.0	115.0	125	135.0	
104	100.0	125.0	26	130	+	Died within 24 hours. 19′ above 40	37	80.0	96.0	17	100.0	
106	98.0	110.0	0	110	+	Died within 24 hours	119	80.0	98.0	18	102.0	47′ above 40
116	90.0	85.0	34	86	–	Died within 24 hours. 15′ above 40	107	78.0	110.0	0	110.0	44′ above 40
							118	78.0	108.0	7	110.0	75′ above 40
Ave.	109.8	102.4		105			Ave.	89.4	103.4		109.2	

TABLE 4

Six per cent gum acacia in 2 per cent $NaHCO_3$, *12 cc. per kilo*

DIED								RECOVERED OF SHOCK						
Serial number	Initial pressure	Pressure end of observation period	Time between observation period and treatment	Treatment		Slight abdominal hemorrhage	Remarks	Serial number	Initial pressure	Pressure end of observation period	Time between observation period and treatment	Treatment		Remarks
				Pressure at beginning	Pressure at end							Pressure at beginning	Pressure at end	
	mm.	*mm.*	*min.*	*mm.*	*mm.*				*mm.*	*mm.*	*min.*	*mm.*	*mm.*	
134	135.0	117.0	13	120.0	135.0	–	Died withing 24 hours. 8′ above 40	130	130.0	120.0	0	120.0	130.0	134′ above 40
126	130.0	93.0	1	93.0	96.0	+	6′ above 40	52	110.0	108.0	0	108.0	120.0	11′ above 40
139	120.0	100.0	0	100.0	110.0	–	77′ above 40	60	105.0	75.0	46	85.0	100.0	
127	120.0	95.0	17	95.0	100.0	–	12′ above 40. Cardiac; clamped 2 hours 15′; clamp partly open ?′	129	100.0	116.0	7	116.0	120.0	134′ above 40
123	110.0	115.0	3	115.0	120.0	+		61	100.0	84.0		85.0	100.0	
58	101.0	100.0		100.0	110.0	–		142	100.0	130.0	41	130.0	140.0	Died end of 3rd day. No autopsy
63	90.0	94.0	43	95.0	103.0	–	Died within 24 hours	141	83.0	87.0	38	90.0	100.0	
143	85.0	108.0	16	110.0	120.0	–		137	80.0	120.0	24	120.0	120.0	56′ above 40. Died end of 3rd day. No autopsy
125	80.0	95.0	2	95.0	106.0	+		50	80.0	85.0	0	85.0	110.0	30′ above 40
								133	75.0	97.0	21	100.0	110.0	
								57	70.0	94.0	21	98.0	98.0	16′ above 40
Ave.	107.9	101.9		102.6	111.1			Ave.	93.9	101.5		103.7	113.4	

TABLE 5

Twenty-five per cent gum acacia (Na), *5 cc. per kilo and 5 per cent* $NaHCO_3$, *5 cc. per kilo*

DIED								RECOVERED OF SHOCK						
Serial number	Initial pressure	Pressure end of observation period	Time between observation period and treatment	Treatment		Slight abdominal hemorrhage	Remarks	Serial number	Initial pressure	Pressure end of observation period	Time between observation period and treatment	Treatment		Remarks
				Pressure at beginning	Pressure at end							Pressure at beginning	Pressure at end	
	mm.	*mm.*	*min.*	*mm.*	*mm.*				*mm.*	*mm.*	*min.*	*mm.*	*mm.*	
175	125.0	111.0	14	115.0	127	–	13′ above 40. Died within 24 hours	166	100.0	102.0	0	102.0	109	
176	114.0	129.0	4	130.0	150	–		182	98.0	117.0	8	118.0	125	
170	110.0	95.0	9	95.0	120	+	Died within 24 hours	165	95.0	112.0	0	112.0	121	
41*	105.0	91.0	14	92.0	100	+	Died within 24 hours	172	95.0	110.0	32	112.0	124	Died end of 3rd day. No autopsy
159	100.0	112.0	51	112.0	120	–	Died within 24 hours	171	90.0	109.0	34	114.0	115	129′ above 40
169	95.0	112.0	26	117.0	124	–	Died within 24 hours	163	60.0	107.0	20	107.0	117	60′ above 40
49†	90.0	83.0	3	85.0	100	–	Died within 24 hours	43		100.0	9	100.0	115	70′ above 40
164	75.0	76.0	22	76.0	82	–	Died within 24 hours							
174	75.0	75.0	8	76.0	94	–	Died within 24 hours. Cardiac; clamped 2 hours 15′, partly open 5′							
Ave.	98.8	98.3		99.8	113			Ave.	89.7	108.1		109.3	118	

* 4 per cent $NaHCO_3$ combined with acacia. Some blood removed for Hb. determination.

† 5 per cent $NaHCO_3$ combined with acacia.

Cardiac cases. A certain number of animals die of, or are threatened by, cardiac failure during the period of occlusion. We have not had an opportunity to study this condition carefully. It comes on as a rule during the first hour of the clamping period. Its imminence is indicated by an irregularity in the force and rhythm of the pulse as registered by the arterial manometer. If, when this is noted, the clamp on the cava is cautiously opened and the heart massaged through the thorax, the pulse in the course of a few minutes may become perfectly regular again. Once the heart has thus recovered, the arterial pressure often can again be held down by the clamp for the rest of the clamping period and the heart, as a rule, will not again become irregular. But quite as often opening the clamp and allowing the pressure to rise at the time cardiac failure becomes imminent does not help matters. Sometimes, indeed, it seems to do harm: the irregularity increases, the pressure falls and the heart stops.

The further course of the cases showing these cardiac disturbances, but which, through temporary opening of the clamp, can be carried to the period of observation, seems to indicate, however, either that the damage that led to the cardiac disturbance was not altogether transient, or that the effect upon the heart is a measure of the damage the occlusion of the cava has inflicted on the body generally. If we include in this analysis only those cases in which it was possible to carry out the clamping stage as usual, with the exception of the few minutes during which it was necessary to open the clamp in order to save the heart, and exclude the cases of marked abdominal hemorrhage, the whole series contains 22 cardiac cases. In 10 or 11 of these cases the arterial pressure after removing the clamp began to fall before the 2-hour period of observation had elapsed. In accordance with the rule, all of these died excepting the one of this character previously referred to. Twelve of the cases belong, therefore, to the series from which we deduce the results of applying measured damage. Six of these died within 48 hours despite the fact that their clamping period is usually somewhat curtailed by the period during which the heart is being given a chance to recover.

That the cardiac cases are somewhat handicapped is indicated also by the behavior of the arterial pressure during the observation period. While the average arterial pressure of all of the cases that died of clamping the cava was 100.1 mm. Hg. at the close of the 2-hour period, of all that recovered, 106.3 mm. Hg., the average of the 6 cardiac cases that died was 85.2, of the 6 that recovered, 100.7. This last group

includes one case (see table 6) whose arterial pressure rose to 135 mm. Hg., the highest pressure reached after 2 hours in the whole series of 219 animals. If we exclude this exceptional case, the average pressure of the cardiac cases that recovered becomes 93.8 mm. Hg. These cases, therefore, have to contend with a circulation that is inferior to that of the general run to a degree that is indicated by a lower arterial pressure amounting to from 12 to 15 mm. Hg.

It seems only fair that these facts should be taken into account in considering the effects of treatment. With this end in view, we have included in the statistics the figures obtained after excluding the cardiac cases that died. It will be seen, however, that the general relation of the several groups to each other is not materially altered by this method of presenting the results.

THEORY OF THE TREATMENTS

The object we had in mind when this investigation was begun was to determine the relative efficacy of solutions containing gum acacia and a crystalloid in the prevention of death due to trauma. The number of solutions that might have been studied in such an investigation was almost limitless. Inasmuch as there were reasons for reaching an early decision, and inasmuch as it was realized that large numbers of animals would be required to satisfactorily test each solution, it was necessary to reduce the number tested to the smallest that might suffice to indicate the advantages and disadvantages of the different types of combinations.

At the time, a solution containing bicarbonate as the crystalloid had been recommended for use at the front by the English Committee on Shock and Allied States (13). By starting the tests with this first Bayliss solution, therefore, we felt it would be possible at one and the same time to test the claims made for this solution and to determine the efficacy not alone of a solution practically isotonic as regards both colloid and crystalloid but also one containing an alkali which also was being recommended for use in the treatment of shock (14). The results obtained with such a solution could then be compared with the results obtained with a hypertonic solution of the same ingredients. For reasons that will become evident, solutions containing glucose instead of bicarbonate were also tested.

1. Gum acacia, 6 per cent, in $NaHCO_3$, 2 per cent. This solution was made up about as described in the private report of the Special

TABLE 6

Twenty-five per cent gum acacia (Ca) *5 cc. per kilo and 18 per cent glucose, 5cc. per kilo*

DIED								RECOVERED OF SHOCK						
Serial number	Initial pressure	Pressure end of observation period	Time between observation period and treatment	Treatment: Pressure at beginning	Treatment: Pressure at end	Slight abdominal hemorrhage	Remarks	Serial number	Initial pressure	Pressure end of observation period	Time between observation period and treatment	Treatment: Pressure at beginning	Treatment: Pressure at end	Remarks
	mm.	*mm.*	*min.*	*mm.*	*mm.*				*mm.*	*mm.*	*min.*	*mm.*	*mm.*	
150	130.0	90.0	19	94.0	122.0	+	5′ above 40. Cardiac; clamped 2 hours 16′, clamp partly open 19′	153	125.0	127.0	30	132.0	150.0	
90	125.0	107.0	6	107.0	130.0	−	10′ above 40	72	120.0	118.0	2	118.0	135.0	38′ above 40
79	116.0	99.0	10	100.0	118.0	−		77	120.0	93.0	5	96.0	118.0	Died end of 3rd day. Peritonitis; cardiac; clamped 2 hours 15′, clamp partly open ?′
64	115.0	84.0	25	87.0	90.0	+	25′ above 40	54	105.0	90.0	28	97.0	105.0	Died end of 3rd day. No autopsy
89	110.0	105.0	10	105.0	118.0	+	Died within 24 hours	69	105.0	118.0	13	120.0	137.0	
76	100.0	108.0	22	110.0	120.0	+		75	104.0	130.0	12	130.0	130.0	10′ above 40
156	100.0	114.0	14	115.0	128.0	−	134′ above 40	136	100.0	130.0	28	135.0	165.0	
73	85.0	95.0	27	100.0	118.0	−		147	100.0	108.0	4	108.0	125.0	
84	78.0	75.0	24	78.0	92.0	−	Cardiac; clamped 2 hours 20′, clamp partly open 5′	80	98.0	98.0	6	98.0	120.0	11′ above 40
								155	85.0	124.0	22	124.0	128.0	
								88	75 0	103.0	9	108.0	130.0	Died in 51½ hours. Peritonitis; cardiac; clamped 2 hours 20′, clamp partly open 5′
Ave.	106.6	97.5		99.6	115.1			Ave.	103.4	112.6		115.1	131.2	

Committee on Shock and Allied Conditions of November 9, 1917. Enough sodium bicarbonate to make a 2 per cent solution was added to a 6 per cent solution of gum acacia in water. The solution was heated and the precipitated $CaCO_3$ centrifuged off. The solution was then heated again, and if another precipitate formed, this again was removed, this process continuing until heating the mixture sufficiently long to sterilize it no longer gave rise to a precipitate.

The only essential divergence from the method of preparation recommended by the English Committee consisted in not conducting the heating of the solution in a sealed vessel. Our method of preparation probably converted most of the bicarbonate into carbonate. In this respect, however, the difference between the two methods is only relative. For even admitting that in the process of preparation as recommended by the English Committee the bicarbonate is dissociated only to the extent that occurs ordinarily at room temperature and in atmospheric air, only a certain proportion of it would be present as such (15), and the dissociation would tend to go further at the time the solution, heated for injection, is exposed to atmospheric air. This change in the bicarbonate can not be avoided by sterilizing the dry substance by dry heat, for unless this is done in the presence of CO_2 under tension, the bicarbonate at the sterilizing temperature also would be converted into carbonate practically completely (15). As a matter of fact, unless solutions containing bicarbonate are kept exposed to CO_2 at a relatively high tension at the time they are being injected more or less carbonate will always be present.

In order to understand the action of this solution on the blood volume and on the alkali reserve, it is necessary to know approximately the salt concentration remaining in solution after precipitating the lime and after boiling. The amount of calcium contained in gum acacia seems to vary within very wide limits. If we take the figure given by Bayliss, namely, 2.23 per cent (16), the minimum amount of $NaHCO_3$ left in the 2 per cent solution after adding it to the 6 per cent solution of gum acacia would make a 1.9 per cent solution. If boiling converts all of the bicarbonate into carbonate, the minimum salt concentration would be 1.5 per cent Na_2CO_3. The freezing point of the solution, if the conversion into carbonate were complete, would be about $-0.6°$C. or $-0.77°$C. for the unconverted bicarbonate; in other words the freezing point of the solution as injected would probably be somewhat lower than that of the blood plasma ($-0.56°$C.). When these experiments were done, we were of the opinion that this first

Bayliss solution was isotonic with respect to the blood colloids. We now know that it is slightly hypertonic in this respect also. But the differences from isotonicity are probably not so large as to be of any particular significance (7).

The dose of the solution used in these experiments, 12 cc. per kilo of body weight, is somewhat larger than that (500 cc.) recommended by the English Committee for use in man, which for a 60 kilo man is 8.3 cc. per kilo. The report does not specify the rate at which this dose is to be administered. We used the larger dose because it was about the amount needed to restore the blood of a shocked animal to its normal concentration (7), to say nothing of the amount needed to make good the stagnated blood. The larger dose was used also in order that the alkali would come closer to the amount needed to combat the average acidosis that develops in experimental shock. But even in the larger dosage the alkali, presumably, was not nearly sufficient for this purpose (7). The time taken to administer our dose averaged 29 minutes.

2. Gum acacia, 25 per cent, and sodium bicarbonate, 5 per cent. The solutions for this treatment were made up and administered in several ways. At first enough $NaHCO_3$ to make a 5 per cent solution was dissolved in the 25 per cent gum, the solution heated and the precipitate centrifuged off. The gum, however, turned deep brown in color, especially during sterilization, and while we had no evidence that the colored gum was harmful, it was deemed best not to use it. The next method consisted in adding a very slight excess of the bicarbonate to the gum solution, applying heat and then centrifuging off the precipitated $CaCO_3$. The solution was then neutralized carefully with HCl and sterilized in sealed centrifuge tubes. If, during sterilization, a new precipitate formed this was thrown down in the centrifuge. This solution of sodium acacia was decanted into a sterile burette as needed and the bicarbonate, dry sterilized, added in sufficient amount to make a 5 per cent solution of bicarbonate. In by far the largest number of experiments, 5 cc. per kilo of the sterile, neutralized sodium acacia, made as described above, were first given and immediately followed by a solution made by adding dry-sterilized bicarbonate to sterile water in sufficient amount to make a 5 per cent solution of $NaHCO_3$. From time to time the proportions and the dosage were varied (see table 5) but never by enough to materially alter the conditions as here described. The administration of the acacia solution

occupied about a half-hour, of the carbonate solution about 23 minutes. The rate of injection of the solution, calculated as bicarbonate, was therefore 0.65 gram per kilo and hour. The rate recommended by the English Committee (17) was about 0.95 gram per kilo and hour in the form of a 4 per cent solution of bicarbonate made up, presumably as we have made our solution.

The dosage we employed was based upon the view (16) that 7 per cent gum acacia was isotonic with respect to the colloids of the blood and that, therefore, the water the colloid theoretically could hold would be about the amount needed to restore the blood volume of shocked animals to normal. Newer estimations of the osmotic pressure of gum acacia (4), (7) indicate, however, that the figure used by us was somewhat too low. The bicarbonate was added in sufficient amount to draw into the circulation approximately the amount of water the gum on the basis of the available osmotic data was capable of holding. The amount of alkali administered was greater in the proportion of 25 to 21 than in the dose of the solution consisting of 6 per cent gum and 2 per cent bicarbonate. In neither case, though, was it present in amounts large enough to balance, by chemical means, at least, the acidosis usually present at the time the injection is begun (7).

3. Gum acacia, 25 per cent, and glucose, 18 per cent, were given in two ways. In one series the 25 per cent gum acacia was given first and was followed immediately by the 18 per cent glucose. The carefully filtered gum solution was sterilized under pressure in stoppered centrifuge tubes and in case a turbidity developed the solution was cleared in the centrifuge. In order to avoid the change in the glucose solution that is produced by sterilization at high temperatures, as indicated by the yellow color that develops, the glucose solutions at first were sterilized by pasteurization. Subsequent experience showed, however, that the autoclaved glucose is not injurious; therefore, the latter method of sterilization was finally adopted. The dose of each of the solutions was 5 cc. per kilo of body weight. The injection of the gum again occupied about 30 minutes. The glucose injection averaged about 19 minutes. The latter, therefore, was given faster than the tolerant rate of 5 cc. per kilo and hour. The rationale of this dosage was the same as that underlying the acacia-carbonate dosage. The acacia was in such a concentration that through its osmotic tension it could easily hold the water which the 18 per cent solution of glucose would draw into the circulation. The effect of

TABLE 7

Twenty-five per cent gum acacia (Ca) *in 18 per cent glucose, 5 cc. per kilo and hour*

DIED								RECOVERED OF SHOCK						
Serial number	Initial pressure	Pressure end of observation period	Time between observation period and treatment	Treatment: Pressure at beginning	Treatment: Pressure at end	Slight abdominal hemorrhage	Remarks	Serial number	Initial pressure	Pressure end of observation period	Time between observation period and treatment	Treatment: Pressure at beginning	Treatment: Pressure at end	Remarks
	mm.	*mm.*	*min.*	*mm.*	*mm.*				*mm.*	*mm.*	*min.*	*mm.*	*mm.*	
198	115	85.0	18	85	95	++	4′ above 40. Cardiac; clamp partially open 13′	211.0	145.0	135	26	138.0	135.0	Cardiac; clamp partially open ?′
201	115	106.0	35	109	120	+	Died in 8 hours. 7′ above 40	200.0	130.0	110	0	110.0	122.0	5′ above 40
197	100	100.0	18	105	130	+	Died within 24 hours	209.0	125.0	125	5	125.0	150.0	113′ above 40
199	90	102.0	9	107	120			191.0	115.0	92	12	94.0	95.0	Lost 10 cc. blood
205	90	108.0	18	109	125		Died within 24 hours. Chronic liver disease. Pleura full of bloody exudate	212.0	115.0	100	0	100.0	115.0	46′ above 40. Cardiac, clamped 2 hours. 16′; partly open ?′
								189.0	110.0	115	20	115.0	125.0	3′ above 40. Died end of 3rd day; slight peritonitis
								193.0	110.0	130	8	130.0	135.0	131′ above 40
								202.0	105.0	108	0	108.0	125.0	3′ above 40. Died in 3 days; peritonitis
								187.0	100.0	125	25	125.0	120.0	91′ above 40
								190.0	95.0	112	0	112.0	125.0	
								186.0	90.0	85	0	85.0	100.0	Cardiac; clamp partly open ?′

								213.0	90.0	91	13	95.0	110.0	
								183.0	88.0	105	12	100.0	110.0	36′ above 40. Ether given during treatment
								185.0	85.0	80	25	85.0	95.0	
								217.0	85.0	92	18	97.0	120.0	
								214.0	80.0	90	0	90.0	100.0	
Ave.	102	100.2		103	118			Ave.	104.2	106		106.8	117.6	
215	75	64	17	60	85		Died within 24 hours. Cardiac							

this combination upon the blood volume of shocked animals has not been determined. It has, however, been shown in normal animals that the results obtained with it are in general accord with theory (7).

4. Gum acacia, 25 per cent, and glucose, 18 per cent, in combination. In the foregoing series the acacia and glucose were given in the succession, in the dosage and at the rates described in order to make the conditions comparable with those obtaining in the series that received 25 per cent gum and 5 per cent bicarbonate solutions. The two latter substances could not conveniently be given in one solution because, as has been said, the gum acacia in strongly alkaline solution seemed to be considerably changed during sterilization. And it seemed advisable to give the aftercoming crystalloid as rapidly as was safe in order to bring rapidly into the circulation the water needed to dilute the gum acacia. There were reasons for suspecting, however, that the combined administration of acacia and crystalloid might have some advantages over their successive administration. For until this water was added, the circulation of blood would be slowed by the increase in its viscosity produced by the strong gum solution. By combining the glucose with the gum this necessity no longer existed and it then was possible to inject the glucose at a slower rate, one which in normal animals does not lead to glycosuria (18). In this series of experiments, therefore, the gum and the glucose were combined in one and the same solution. The method of making up this solution has been described in another place (19). It was injected at the rate of 5 cc. per kilo and hour; the glucose, therefore, entered the circulation at the subtolerant rate of 0.9 gram per kilo and hour.

The effect of the injection of solutions 3 and 4 upon the alkali reserve has not been determined. According to Macleod and Hoover (20), alkaline glucose injections increase the amount of lactic acid in the blood. Acid glucose solutions, they showed, do not have this effect. The present solution, through the reaction of the acacia, is faintly acid: presumably, therefore, there would be no acid formed through the process described by Macleod and Hoover.

STATISTICS

Excluding, as has been explained, the cases in which the arterial pressure began to fall consistently before the close of the 2-hour period of observation, it is seen (tables 1, 2, 3) that of the 23 acceptable cases composing the control series, 11, or 48 per cent, died within 48 hours,

4 of these, or 17.4 per cent of the group, within 24 hours. In the series treated with the combination of 6 per cent gum acacia and 2 per cent sodium bicarbonate (tables 1, 4) there are 20 acceptable cases of which 9, or 45 per cent, died within 48 hours, and 1 of these, or 5.0 per cent of all the acceptable cases, within 24 hours. In the series receiving in succession 25 per cent gum acacia and 5 per cent sodium bicarbonate (tables 1, 5) there are 16 acceptable cases; 9, or 56 per cent, died within 48 hours; and 8 of these, or 50 per cent of all in the group, died within 24 hours. It was so obvious that this treatment was doing harm that it was not regarded as necessary to accumulate as many cases as we have in the other groups. The group receiving 25 per cent gum acacia and 18 per cent glucose in succession (tables 1 and 6) comprises 20 acceptable cases, 9 of which, or 45 per cent, died within 48 hours, and of these 1, or 5.0 per cent of all the cases, within 24 hours. And, finally, the series receiving the gum and glucose in combination (tables 1 and 7) contains 21 acceptable cases: 5 of these, or 24 per cent, died within 48 hours, and 3 of these, or 14.3 per cent of the whole number, within 24 hours.

The two most obvious results coming of this comparison of the several groups are *a*, the unfavorable showing of the series receiving the 25 per cent gum acacia and 5 per cent sodium bicarbonate in succession, as regards both total mortality and duration of life of the fatal cases; and *b*, the favorable showing of the series receiving 25 per cent gum in combination with 18 per cent glucose as regards total deaths. In the matter of one day deaths, the latter series is only slightly better than the control series and is not so favorable as the series that received the 6 per cent gum in 2 per cent bicarbonate or 25 per cent gum and 18 per cent glucose. The number of one day deaths in these three series is so small, however, that a difference of one case in either direction would practically have eliminated all differences. The high 24 hour mortality of the series receiving 25 per cent gum and 5 per cent bicarbonate cannot, however, be accounted for in this way. This treatment unquestionably does harm.

As has been said, a high arterial pressure at the end of the observation period seems to favor recovery. The average pressures of all of the groups at this time (table 2) are, however, so nearly alike that it is not necessary to make any allowance for such effect as this factor may have.

From the cases that died and are otherwise acceptable have been excluded, as has been explained above, the cases in which considerable

abdominal hemorrhage is found at autopsy. The necessity for excluding such cases is obvious. On the other hand, as has been said, the decision as to whether the hemorrhage is marked or slight rests upon a judgment which often it is difficult to make. In order to take this into account we give in columns 6, 7, 8 and 9 of table 1 the statistics as obtained by excluding all of the fatal cases in which at autopsy even a small amount of bloody fluid was found in the abdomen. It is seen that the relative number of hemorrhage cases in each group is practically the same, demonstrating again its accidental origin. It follows that when they are excluded the relative mortality rate is not markedly affected (column 9); the differences already presented (column 5) are merely somewhat emphasized.

In columns 10 and 11 (table 1) it is seen that with the exception of two groups, practically all of the deaths occurring within 24 hours were complicated by slight hemorrhage. The exceptions are in the case of the groups receiving bicarbonate and seem to emphasize from still another aspect the deleterious effects of alkali. Set over against this is the fact that all of the one day deaths occurring in the series receiving 25 per cent gum acacia in 18 per cent glucose were complicated by the slight hemorrhage; there are left in this group altogether only 1 or 2 deaths if those with slight hemorrhage are excluded.

It has been pointed out that, in general, cases with high initial arterial pressures are more apt to die than cases with low initial pressures. On this basis it is seen (table 2, columns 1 and 9) that the group with the best initial chance, namely, those treated with 25 per cent gum acacia and 5 per cent sodium bicarbonate, actually is the one that did the worst; while the group with the worst chances in this respect, namely, the one receiving 25 per cent gum acacia followed by 18 per cent glucose, did quite as well as any; and that the treatment that gave the best results, namely, the 25 per cent gum acacia in 18 per cent glucose, was tried out on a group whose initial chances were not particularly favorable.

The results obtained by excluding, for reasons already given, the cases in which the pressure failed to fall to 40 mm. Hg. within 30 minutes after applying the clamp to the cava and lived are seen in columns 15, 16, 17 of table 1. These exclusions cause no important changes in the relative positions of the several groups. The treatment consisting of 25 per cent gum acacia and 5 per cent sodium bicarbonate still gives the worst result; that consisting of 25 per cent gum acacia in 18 per cent glucose the best. The group receiving 6 per cent gum

acacia in 2 per cent sodium bicarbonate gains a little, that receiving 25 per cent gum acacia followed by 18 per cent glucose, somewhat more, on the control group.

Finally, we take into consideration the handicap of the cases in which the heart during the clamping period threatened to fail, by calculating the results after excluding those of these cases that die. Columns 12, 13, 14 (table 1) show that this precaution also changes the results but little. Its main effect is to improve slightly the results of the administration of the solutions containing glucose.

It has already been stated that the series of acceptable cases includes some 12 so-called heart cases, and that of these 6 recovered of shock. It is interesting to add here that of the 6 that recovered, 5 were in the series that were treated with glucose, namely, 2 in the group receiving acacia and glucose in succession, and 3 in the group receiving the solutions simultaneously. One was in the control group. While the number of cardiac cases is not sufficiently large to permit of any definite conclusion, it certainly is suggestive that with but one exception, all of the cardiac cases that recovered of shock had received intravenously a substance, glucose, which is known to affect favorably the contraction of heart muscle.

Thus far we have confined ourselves to the action of the solutions on the state that develops, to all intents and purposes, immediately after unclamping the vena cava, that is, on shock. It has already been stated that 8 of the animals that had shown marked improvement from this first state died rather unexpectedly, for the most part, toward the end of the third day. The possible significance of such deaths did not at first occur to us and as a consequence we have autopsy notes on only four of the cases. These four presented signs of peritonitis ranging from a fibrinous deposit on the peritoneum to a sanguinopurulent exudate. The fatal issue in these instances, therefore, can be attributed neither to shock nor to a late deleterious action of the injected solutions. The other four cases not autopsied were scattered among three of the groups. In view of the relatively small number of these cases—4 out of the 100 that were acceptable—and in view of the regular presence of peritonitis in the four cases that came to autopsy, and in view, furthermore, of the many other possible causes of death, even if peritonitis were excluded, it certainly seems unnecessary to attribute these deaths to a delayed action of the injected solutions. It might be added that the incidence of peritonitis was not high when it is taken into account that the peritoneum was open for

2¼ hours at a time when its resistance, owing to the slowing of the circulation, must have been quite low, and that there was the possibility of infection through the walls of the gastro-intestinal tract damaged by the long-lasting ischemia.

DISCUSSION

The foregoing analysis shows that no matter how the statistics are viewed, the relative efficacy of the solutions tried is − 25 per cent calcium acacia in 18 per cent glucose > 25 per cent gum acacia followed by 18 per cent glucose = or > 6 per cent sodium acacia in 2 per cent sodium bicarbonate = or > untreated controls > 25 per cent sodium acacia followed by 5 per cent sodium bicarbonate.

In attempting to account for these results it should first be recalled that in shocked animals all of the solutions that have been tested are capable of restoring the blood volume and the blood pressure and of maintaining these to practically the same degree under similar circumstances. Considering the end in view it certainly seems justifiable to assume that these responses are desirable. The fact, therefore, that some of the solutions, 6 per cent gum acacia in 2 per cent sodium bicarbonate and 25 per cent gum acacia followed by 18 per cent glucose, for example, effect but little, if any, reduction in the death rate of traumatized animals, may mean that these solutions are not entirely innocuous and that the harm they do just balances the good that results from the improvement they effect in the circulation. There can, of course, be no doubt but that the treatment in which the 25 per cent sodium acacia is followed by 5 per cent sodium bicarbonate is harmful. Is it possible to ascertain from the limited data available wherein this harmfulness consists?

Gum acacia in 6 per cent solution we believe is wholly innocuous. This is proved by our own experience. Even a 25 per cent solution of gum acacia in the dosage which we have used is quite innocuous to normal animals. When such a solution is given rapidly to dogs until the blood plasma becomes a 10 per cent solution of gum acacia no obvious symptoms develop (22). Czerny (23) found the maximal non-lethal dose for cats and rabbits to be 4.66 grams per kilo. This is almost four times the amount contained in the dose we give in shock and in the concentration (24 per cent) used by Czerny, would convert the plasma of these animals into an 8 per cent solution of gum. And Czerny did not even use aseptic precautions. The innocuousness of the hypertonic

solution is illustrated also by the biological tests we have been making of the solution consisting of 25 per cent gum in 18 per cent glucose made up for the treatment of shock in man (19). Each batch of this solution is tested by injecting in the course of a half-hour 10.0 cc. per kilo into an animal previously bled to the extent of more than 3 per cent of its body weight. In none of the animals so treated has there been the slightest evidence of harm.

There is no question, though, but that concentrated gum in the dosages we have employed may do harm if it is injected *rapidly* into an animal about to die of shock. Under these circumstances, in our experience, the heart occasionally has stopped as though it had fibrilated. Presumably, this is the result of the slowing of the circulation that comes of the high viscosity of the injected gum solution. At any rate this harmful action cannot be attributed to the osmotic properties of the gum, for nothing of the kind is seen when the gum in equally hypertonic solution is injected more slowly, not even when it is in combination with hypertonic glucose. Nor can it be due to any chemical action of the gum, for inasmuch as the injected gum remains for some time in circulation, there is finally just as much of it in circulation when it is injected slowly as when it is injected rapidly; yet, under the former circumstances stoppage of the heart does not occur. To be sure, calcium acacia has been used almost exclusively in these tests; but as regards osmotic (4) and viscosity (16) factors, as these affect the blood, the sodium and calcium acacias are presumably practically alike. In the course of our experiments we have given large doses of both of these acacias and have not been able to satisfy ourselves that there is any difference in their action. Furthermore, if sodium acacia acts deleteriously through chemical means, the results obtained from the injection of the sodim acacia in combination with isotonic bicarbonate should have been almost as bad as the results of the injection of the 25 per cent solution followed by the hypertonic carbonate, for the amount of gum given with the former (0.72 gm. per kilo) is not very much smaller than the amount given with the latter (1.25 gm. per kilo). The difference in the results following the injection of these two solutions, though, is quite striking.

The result of this analysis, as far as it has gone, is to indicate, therefore, that it is neither the sodium acacia *per se* that does the harm nor its hypertonicity, but rather the alkali that is injected with or after it, to a certain extent also the high viscosity of the more concentrated gum solution that lasts until it is diluted by its own osmotic action

and by that of the crystalloid subsequently injected. It is not necessary to conclude at this juncture, however, that the bicarbonate is injurious by virtue of its alkalinity. It may be that saline crystalloids are undesirable in the treatment of shock, possibly because they accumulate and disturb the salt balance of the body.

It was considerations of this character that led us, when it was found that the gum in combination with bicarbonate is without value in the treatment of shock, to try out glucose as the crystalloid, especially in view of the indications obtained by one of us in collaboration with Woodyatt (24) that it ameliorates the symptoms of experimental shock better than do salines.

The series of experiments in which the glucose replaced the bicarbonate but comparable in every other respect save the nature of the gum, clearly demonstrated the superiority of the glucose as a means of drawing the water to the gum. Yet this succession of gum and glucose was without any marked effect upon the death rate due to the measured trauma. The next step was to eliminate the period of increased viscosity of the blood by injecting the gum and the crystalloid simultaneously. It was by this method that the maximum saving of life was effected.

It is conceivable that this beneficial action is due not alone to the osmotic action of the glucose but also to some specific action it may exert. With regard to the relative merits of salt and glucose as a means of bringing water into the circulation, it may first be pointed out that there is relatively little reason, other than osmotic, for administering salts in shock, for there is, so far as is known, no salt deficit in that state. Neither is there any lack of glucose (14). But glucose has the advantage over most other crystalloids that might be used for this purpose, that through oxidation and polymerization, the organism can quickly put it out of the way after it has accomplished the purpose of bringing water to the gum acacia. However this may be, there is abundant evidence in the literature indicating that glucose of itself acts beneficially in clinical shock and allied conditions. Woodyatt, with Sansum and Wilder, obtained favorable results from sustained injection of glucose in hypertonic solution in two cases having features of shock (24). Hypertonic glucose has been found to be of benefit in pneumonia (25) and in a number of other conditions.

There are a number of ways in which glucose might prove of functional value to the traumatized organism. The fact that in shock there is no deficiency of blood sugar does not necessarily mean that

in this respect the administration of glucose would prove futile. The beneficial results said to come of feeding sugar to soldiers on the march (26), for instance, can not be attributed to any deficiency of glucose in the blood (27). Whatever the mechanism of the relief from fatigue may be, it seems likely that the same or a similar mechanism might work to relieve shock when glucose is administered. It is well known that glucose in excess of the normal blood content is oxidized in the tissues (28), and that it is used as a food when it is introduced into the system parenterally (29). Glucose also improves and sustains the beat of the perfused heart (30), (31), as well as the contractions of other types of muscle (32), (33). In this connection it is of interest to recall that of the six cardiac cases that recovered of shock, five were in the groups that received glucose.

Over and above all of the considerations that have thus far been discussed, there was another which held out hopes that hypertonic solutions might prove of value in the treatment of shock. Eyster and Wilde (34) have found that certain crystalloids in hypertonic solution, glucose among others, cause an immediate increase in cardiac output and a vasodilatation. These actions apparently are independent of the resultant hydremia; indeed, they seem to be specific. But whatever may be the cause, it is obvious that these are exactly the responses best calculated to improve the circulation in shock.

It will be noted that we have not tested the efficacy of blood in the treatment of shock. One reason for not doing so was the number of animals that would have been required, while another was the fact that our animals were not suffering from any loss of blood. When, as in pure shock, there is no actual loss of blood, but rather a concentration of the blood and a crowding of the small veins and capillaries with corpuscles, the introduction of more blood, unless it promptly betters the condition, would seem to be contra-indicated. At least this is the inference one is led to draw from the effects of the injection of blood into normal animals. Starling points out (35) that under these circumstances the blood fluids do not remain long in the vessels but pass into the lymphatics, leaving behind the corpuscles and a certain proportion of the proteins of the plasma. This concentration of the blood raises its viscosity and tends to embarrass the circulation; there is produced a state of affairs similar to the one we are trying to combat in shock. For this reason it is conceivable that blood plasma may have some advantages over whole blood in the treatment of non-hemorrhagic shock. Furthermore, the injection of blood even in suit-

able cases is by no means entirely free of danger (36). It is, therefore, of interest to recall in this connection that blood transfusion is not essential to recovery even from the severest acute hemorrhage, if only the blood bulk can be restored in other ways (37). The present investigation possibly indicates that a restoration of blood volume in which the tissues are made to participate through osmotic action is more efficacious than one effected through the injection of the full amount of fluid needed to make good the reduction.

Results in man. After we had convinced ourselves through the animal experiments described in this paper that hypertonic gum and glucose are not alone innocuous if given slowly, but actually save a certain number of animals from death by trauma, we began the use of this solution in the treatment of shock in man. The results obtained in eleven cases have been recently described in another place (19). It need only be stated here that they fully confirm the conclusions reached in this paper.

SUMMARY

Of animals traumatized by holding the arterial pressure down to 40 mm. Hg. for 2 hours and 15 minutes by partially occluding the inferior vena cava— 48 per cent die within 48 hours.

When treated with:

a. 6 per cent gum in 2 per cent sodium bicarbonate 12 cc. per kilo of body weight 45 per cent die within 48 hours.

b. 25 per cent gum followed by 5 per cent sodium bicarbonate, of each 5 cc. per kilo of body weight 56 per cent die within 48 hours.

c. 25 per cent gum followed by 18 per cent glucose, of each 5 cc. per kilo of body weight 45 per cent die within 48 hours.

d. 25 per cent gum in 18 per cent glucose, 5 cc. per kilo of body weight and hour 24 per cent die within 48 hours.

Not only is the death rate increased by treatment *b*, but death occurs earlier.

These results are taken to indicate that bicarbonate and the high viscosity of a strong gum solution are somewhat harmful, at least, in

traumatized animals; that the harmfulness of the strong, viscid gum can be avoided in part through the osmotic action of hypertonic glucose subsequently injected, but not by bicarbonate; and that when the hypertonic gum and the hypertonic glucose are given simultaneously and slowly so as to avoid altogether the period during which the high viscosity of the gum is hampering the circulation, a maximum saving of life can be effected. The beneficial results presumably are due to the internal tranfusion (38) effected by the hypertonic solutions, to the maintenance of the increased blood volume through the colloidal and possibly other properties of the gum acacia, to the action of the hypertonic solution on the heart and blood vessels, and to the specific action of glucose on nutrition in general and on that of the heart muscle in particular.

BIBLIOGRAPHY

(1) Erlanger, Gesell and Gasser: This Journal, 1919, xlix, 90.
(2) Erlanger and Gasser: This Journal, 1919, xlix, 151.
(3) Erlanger and Gasser: This Journal, 1919, xlix, 345.
(4) Gasser, Erlanger and Meek: This Journal, 1919, l, 31.
(5) Welch: Allbutt's System of medicine, 1910, vi, 779.
(6) Erlanger, Gesell, Gasser and Elliott: Journ. Amer. Med. Assoc., 1917, lxix, 2089.
(7) Gasser and Erlanger: This Journal, l, 104.
(8) Mann: Journ. Amer. Med. Assoc., 1918, lxxi, 1184.
(9) Wiggers and Edwards: Report to Committee on Shock of the National Research Council, July 9, 1918.
(10) Almeida and Almeida: Journ. Amer. Med. Assoc., 1918, lxxi, 1710.
(11) McKibben: This Journal, 1919, xlviii, 331.
(12) Janeway and Jackson: Soc. Exper. Biol. Med., 1915, xii, 193.
(13) Private Rept. of the English Committee on Shock, November 29, 1917.
(14) Cannon: Rept. no. 2, Special Investigation Committee (English), reprinted in Journ. Amer. Med. Assoc., 1918, lxx, 531.
(15) Abegg: Handb. d. anorganisch. Chemie, 1908, ii, 1, 303.
(16) Bayliss: Proc. Roy. Soc., 1916, B lxxxix, 380.
(17) Cannon, Fraser and Cowell: Rept. no. 2, Special Investigation Committee (English), reprinted in Journ. Amer. Med. Assoc., 1918, lxx, 618.
(18) Woodyatt, Sansum and Wilder: Journ. Amer. Med. Assoc., 1915, lxv, 2067.
(19) Erlanger and Gasser: Ann. Surg., 1919, lxviii, 389.
(20) Macleod and Hoover: This Journal, 1917, xlii, 460.
(21) Bayliss: Intravenous injection in shock, London, 1918.
(22) Meek and Gasser: This Journal, 1918, xlv, 548.
(23) Czerny: Arch. f. exper. Path. u. Pharm., 1894, xxxiv, 268.
(24) Erlanger and Woodyatt: Journ. Amer. Med. Assoc., 1917, lxix, 1410.

(25) LITCHFIELD: Journ. Amer. Med. Assoc., 1918, cxxi, 503.
(26) TAYLOR: Digestion and metabolism, 1912, 540.
(27) TAYLOR: Digestion and metabolism, 1912, 256.
(28) MCGUIGAN: This Journal, 1908, xxi, 334.
(29) ZUNTZ AND VON MERING: Arch. f. d. gesammt. Physiol., 1883, xxii, 173.
(30) LOCKE: Journ. Physiol., 1907, xxxvi, 205; also: Zentrabl. f. Physiol., 1914, 78.
(31) EVANS: Journ. Physiol., 1914, xlvii, 407.
(32) NEUKIRCH AND RONA: Arch. f. d. gesammt. Physiol., 1912, cxlviii, 290.
(33) BENEDICT AND CATHCART: Publ. Nutrition Lab. Carnegie Inst., 1913, no. 187, 94.
(34) EYSTER AND WILDE: Journ. Pharm. Exper. Therap., 1910, i, 391.
(35) STARLING: The fluids of the body, 1909, 135.
(36) PEMBERTON: Surg., Gynec., Obst., 1919, xxvii, 276.
(37) ROUS AND WILSON: Journ. Amer. Med. Assoc., 1918, lxx, 219
(38) GESELL: This Journal, 1919, xlvii, 468.

STUDIES IN SECONDARY TRAUMATIC SHOCK

VII. Note on the Action of Hypertonic Gum Acacia and Glucose after Hemorrhage[1]

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The injuries that lead to shock in man almost always are accompanied by more or less hemorrhage. When the hemorrhage is so large as to be dangerous of itself the patient suffers not alone from the immediate and direct consequences of the blood loss but according to prevailing opinion also runs the risk of passing in time into shock properly so-called, possibly, as a consequence of the action of the slowed blood stream upon the peripheral vessels (1).

However this may be, it soon became obvious to us after we had acquired some experience with clinical shock that the surgeon not infrequently is called upon to treat cases with the symptoms of shock in which the extent of hemorrhage is unknown, or in which it is known to be dangerous but for which blood for transfusion is not immediately available. And the question arose in this connection, is it justifiable to employ the hypertonic gum-glucose solution in such instances? Experience gained through the use of the isotonic gum-saline solution in the British Army has led Bayliss to conclude (2) that this substitute for blood is especially useful in those cases in which shock is complicated with a certain amount of hemorrhage. We have not the slightest doubt but that this is true also of the hypertonic gum-glucose solution (3). But the question that concerns us here is not this, but rather, is a hemorrhage that is so extreme as to be apt to prove rapidly fatal as the result of the blood loss itself, a contraindication to the use of the hypertonic gum-glucose solution?

[1] Reported to the Committee on Shock of the National Research Council, July, 1918.

EXPERIMENTAL

The method employed in seeking the answer to this question has been to select two animals as nearly alike as possible as regards weight, vigor and breed, to bleed them both, under ether, to about the same extent in proportion to body weight, and then to give to one of each of the couples 5 cc. of the 25 per cent gum acacia, 18 per cent glucose solution per kilo of body weight in the course of an hour. The usual aseptic precautions were observed. It will be noted in table 1 that the extent of the hemorrhage was not always exactly the same in the two animals of a couple. This came about because of differences in the reaction of the animals to the hemorrhage; the development of threatening symptoms, such as an exceedingly low arterial pressure or marked slowing of the heart rate, indicating that further hemorrhage would probably prove immediately fatal, sometimes forced us to desist in the withdrawal of blood in one or other of the two animals before the intended amount had been drawn.

First hemorrhage. In the dog, according to Fredericq (4), a loss of blood amounting to 2.3 to 4.5 per cent of the body weight is dangerous, the loss of over 4.5 to 5.0 per cent, fatal. Our aim was to remove from the animal by a rapid hemorrhage an amount of blood that would just about prove fatal. As will be seen in table 1, the amount at first

TABLE 1

Showing the amount of blood drawn in per cent of body weight

	GROUP 1		GROUP 2		GROUP 3	
	Dog 1	Dog 2	Dog 3	Dog 4	Dog 5	Dog 6
First hemorrhage	4.41	3.96	4.45	4.64	4.80	4.80
Second hemorrhage	3.16	3.19	3.14	3.13	2.67	2.60
Total	7.57	7.15	7.59	7.77	7.47	7.40

removed ranged between 3.96 and 4.8 per cent of the body weight, yet not one of the animals succumbed, treated or untreated.

In the case of group 1 (dogs 1 and 2, fig. 1, dots) more blood was removed from the test animal (dog 1, large dots) than from its control (dog 2, small dots); the fall in pressure in the case of the control was so profound that the hemorrhage had to be discontinued for fear of immediately killing the animal. This animal had the lower initial pressure. The pressure of the test animal in the course of 1 hour 30 minutes came back to within 10 mm. of its initial value; whereas the pressure of the untreated animal remained 37 mm. Hg. below its normal.

The initial conditions were reversed in the case of the second group (dogs 3 and 4, crosses). For the reasons mentioned above, it was not possible in this case to remove from the test animal (dog 3, large crosses) quite as much blood as from its control (dog 4, small crosses). The difference, though, was inconsiderable. The arterial pressure of the test animal was affected by the hemorrhage much more profoundly than that of the control. The arterial pressure of the treated animal eventually rose to 88 mm. Hg. (the initial was 87); of the untreated animal to 100 mm. Hg. (the initial was 95).

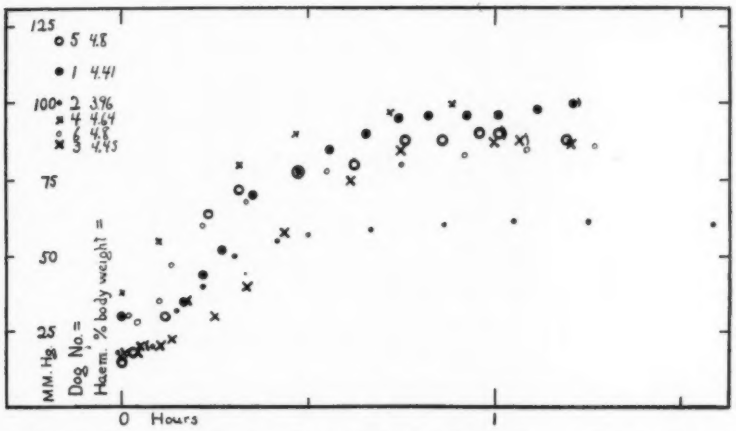

Fig. 1. Chart showing the effects of the first hemorrhage. Dog 1 (treated) ●; dog 2 (control) •; dog 3 (treated) X; dog 4 (control) x; dog 5 (treated) ○; dog 6 (control) ∘. The injection period is the time included in each case between the parentheses.

In the case of the third couple (dogs 5 and 6, circles) the hemorrhages were exactly equal in amount; 4.8 per cent of the body weight was removed. The arterial pressure of the test animal fell lower as the result of the hemorrhage than that of the control. The arterial pressure of the control animal rose to within 4 mm. of the initial level, that of the treated animal to within 30 mm., but the pressure of the treated animal at the end was a bit above that of the untreated animal.

The only conclusion these observations justify is that the injection of the gum-glucose solution did not prejudice the chances of recovery from the effects of a severe hemorrhage. Whether any of the treated

TABLE 2

	GROUP 1				GROUP 2				GROUP 3			
	First hemorrhage		Second hemorrhage		First hemorrhage		Second hemorrhage		First hemorrhage		Second hemorrhage	
	Dog 1, test	Dog 2, control	Dog 1, test	Dog 2, control	Dog 3, test	Dog 4, control	Dog 3, test	Dog 4, control	Dog 5, test	Dog 6, control	Dog 5, test	Dog 6, control
	(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)	(11)	(12)
Initial pressure	110	100	105	85	87	95	90	105	120	90	88	96
Hemorrhage per cent body weight	4.41	3.96	3.16	3.19	4.45	4.64	3.14	3.13	4.8	4.8	2.6	2.6
Pressure at end of hemorrhage	30	18	28	30	18	38	78	74	15	30	76	80
Pressure at or corresponding with beginning of injury	35	34	41	38?	20	45	70	79	18	28	65	77?
Present at or corresponding with end of injury	100	62	95	Dead	88	100?	95	Dead	90	84	78	85
Highest pressure	[100]*	[62]	96	45	[88]	[100]?	95	79	[90]	86	79	88
Pressure last reading	[100]	60	[96]	0	87	[100]?	0	0	88	[86]	73	77

*Bracketed pressure readings are the same as the unbracketed ones next above..

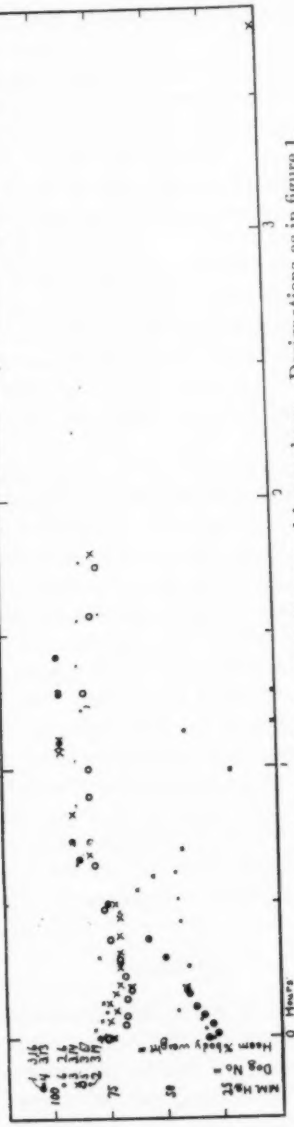

Fig. 2. Chart showing the effects of the second hemorrhage. Designations as in figure 1.

animals would have died had the solution not been administered cannot be determined.

Second hemorrhage. Some two or three days subsequent to the experiments above described each couple was again bled, the amounts taken this time varying between 2.6 per cent and 3.2 per cent of the body weight (see table 1). The amount of blood removed was almost exactly alike in the two animals of each of the couples. The total of the two hemorrhages ranged between 7.15 and 7.77 per cent of the body weight.

In the case of group 1 (dogs 1 and 2, fig. 2, dots) the immediate effect of the hemorrhage was to carry the arterial pressure down to the same level but the absolute drop was greater in the case of test animal. After treatment (dog 1, large dots) the pressure rose to 96 mm. Hg.; this animal recovered. The pressure of the untreated animal rose only to 45 mm. Hg., then fell and the animal died 1 hour 33 minutes after the hemorrhage.

The animals of group 2 (dogs 3 and 4, crosses) were bled to the same extent (3.14 to 3.15 per cent of the body weight). Their pressures as a consequence did not fall very low, but they fell to about the same level. After the hemorrhage for about 36 minutes the pressure of the control animal practically maintained its level while that of the test animal slowly fell. Then the pressure of the control animal began to fall and this animal died 2 hours 20 minutes after the hemorrhage. At the same time under the influence of the injection the decline of the arterial pressure in the treated animal was converted into a rise; it mounted until, by the end of the injection period, it was 5 mm. Hg. above the initial level, but then it began to fall, though slowly, and the animal died 4 hours 30 minutes after the hemorrhage.

Group 3 was bled to the extent of 2.67 per cent (treated) and 2.6 per cent (untreated) of the body weight. The initial pressure of the former was somewhat lower than that of the latter and during and subsequent to the hemorrhage it fell the lower. Both of these animals survived.

DISCUSSION

This analysis of the experimental data shows that the animal of each couple that received the solution did quite as well as, or better than, its control. In thus stating our conclusion we do not desire to give the impression that we are advocating the use of hypertonic gum-glucose solution in the treatment of pure and immediately dangerous hemor-

rhage, though we do believe that when blood is not instantly available it is safe and perhaps advisable to give the gum-glucose solution pending the obtaining of blood. This opinion is based not alone on the results of the present experiments, but also upon our clinical experience (see cases IV, IX and X in our paper on the treatment of shock in man (3)).

Penfield (5) has compared the effect of the following solutions, *a*, 0.9 per cent sodium chloride; *b*, 6.0 per cent gum acacia and 2.0 per cent sodium bicarbonate; *c*, 6.0 per cent gum acacia and 6.0 per cent glucose,—injected into animals after so bleeding them as to hold the arterial pressure down to 40 mm. Hg. for periods ranging between 60 and 89 minutes. The volume of solution injected equalled the amount of blood drawn and varied (as we calculate it) between 4.15 and 5.05 per cent of the body weight. If we except three of his eleven cases, the amount removed and injected averaged 4.44 per cent and did not exceed 4.65 per cent of the body weight. So far as can be determined there were no untreated controls in his series. One of the three of his animals that received the sodium chloride solution died; the average hemorrhage amounted to 4.61 per cent of the body weight. The amount of blood withdrawn at the time of the first hemorrhage in our experiments averaged 4.51 per cent of the body weight and therefore was practically as large as in Penfield's and not a single animal died whether treated or not. There is, therefore, no necessity for concluding that the isotonic solution of sodium chloride in Penfield's hands accomplished any good.

Three of the four animals into which Penfield injected his gum-glucose solution died; the average hemorrhage was 4.29 per cent of the body weight. At this place we desire to correct the statement made by Penfield that he used "gum-glucose solution as recommended by Erlanger." Ours is hypertonic gum-glucose, not isotonic. Having removed this possibility of a misunderstanding, we may say that we are surprised not that three of the four animals treated with the weak gum-glucose solution died, but rather that one of them lived. For Penfield replaced approximately half of the blood in the body with the same amount of a solution which within a very few minutes, through oxidation and polymerization of the glucose, came to consist practically of a solution of 6.0 per cent gum in pure water. The effect that this must have had upon the tissues and the salt balance of the organism, unquestionably accounts for the results Penfield obtained.

The hypertonic gum-glucose solution, of course, is changed in the same way in the organism, but the dose we give, namely, 5 cc. per kilo an hour, is so small, and it is given so slowly, that such disturbance in salt balance as it may cause is negligible in comparison with that caused by the dose of 40 to 50 cc. per kilo employed by Penfield.

CONCLUSIONS

1. The use of hypertonic gum-glucose solution is not contra-indicated in the treatment of shock even when it is complicated by dangerous hemorrhage.

2. The fact that the hypertonic gum-glucose solution does not prejudice the recovery of animals from the effects of a hemorrhage that is apt to result fatally furnishes another proof of the innocuousness of this solution.

BIBLIOGRAPHY

(1) Erlanger and Gasser: This Journal, 1919, xlix, 151.
(2) Bayliss: Intravenous injection in shock, London, 1918.
(3) Erlanger and Gasser: Annals of Surgery, 1919, lxviii, 389.
(4) Fredericq: Travaux du Laboratoire, 1886, i, 133.
(5) Penfield: This Journal, 1919, xlviii, 121.

THE INFLUENCE OF OXYGEN ADMINISTRATION ON THE CONCENTRATION OF THE BLOOD WHICH ACCOMPANIES THE DEVELOPMENT OF LUNG EDEMA

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The enormous and rapid development of edema of the lungs which results from severe gassing of animals with the lung irritants used in warfare offers an unusual opportunity for studying the physiological effects accompanying this pathological condition. The rapidity with which the edema develops precludes the possibility of infection complicating the symptoms observed, and the condition which may develop after exposure to high concentrations of poisonous gas is so severe that the correlated symptoms can hardly be overlooked.

Loss of water from the blood is one of the most characteristic phenomena accompanying the development of edema of the lungs in animals gassed with lung irritants. A concentration of the blood becomes evident at about the time when the edema of the lungs can be first demonstrated. Thereafter the loss of water from the blood and the increase in severity of the edema run roughly parallel. The conclusion was therefore made that the two are interrelated and that the pouring of water into the lungs is the cause of the concentration of the blood.

Other considerations however make it necessary to proceed with caution before accepting this hypothesis. During the acute period after gassing there develops a deficiency of oxygen carried by the blood. Probably due to the poor aeration of the blood in the damaged lung the oxyen content of arterial and venous blood may drop to levels much below normal. The transport of oxygen to the tissues may be still further reduced by the decreased rate of blood flow with the probable result that the oxygenation of the tissues is seriously interfered with.

[1] Published with the permission of the American and British military authorities.

Physiologists have shown that muscle tissue imbibes water when supplied with insufficient oxygen. Based on this observation the hypothesis may be presented that the concentration of the blood is due, not primarily to the development of lung edema, but to the imbibition of water from the blood by the tissues which are not sufficiently oxygenated. To throw some light on the validity of this hypothesis the experiments reported below were carried out.

Goats were gassed with lethal concentrations of chlorpicrin. As soon as possible after gassing, half of the animals were fitted with masks and given oxygen continuously in known quantities by means of a Haldane oxygen apparatus.[2] The other animals were used as controls.

The hemoglobin content was used as an index of the concentration of the blood. Hemoglobin determinations were made frequently using blood obtained by pricking an ear vein. Blood from the heart punctures was also used. The Haldane method was used for the hemoglobin determinations.

When the concentration of the blood was sufficiently marked in the animals to which oxygen was being administered, heart punctures were made and the percentage saturation of the hemoglobin of the bloods from the right and left hearts was determined by means of Barcroft's differential blood gas apparatus.[3] Some difficulty was experienced in obtaining blood from the hearts of animals in which the lungs were large and edematous but the sample was considered satisfactory when it was obtained quickly and with little struggling on the part of the animal.

The protocols are given below as well as curves showing the concentration of the blood. In the curves, the percentage variations from the normal are plotted to make all of the curves directly comparable.

The maximum concentrations observed in the control animals varied from 30 per cent to 60 per cent above normal (average 43 per cent) while in the animals receiving oxygen the variation was from 28 per cent to 75 per cent above normal (average 48 per cent). It is apparent that, on the whole, the blood of animals which received oxygen con-

[2] The Haldane oxygen apparatus furnishes oxygen to a mask fitted with valves for incoming and outgoing air. When the mask is worn by the animal, breathing is easy and the air in the mask may be enriched by varying amounts of oxygen.

[3] We are indebted to Mr. Barcroft, Captain Dunn and Captain Peters for these data.

TABLE I

Chlorpicrin 1/8500 for 25 minutes (10.10 to 10.35 a.m.), August 19, 1918

GOAT 4537	GOAT 4567
9.35 a.m. Hb. 80	10.00 a.m. Hb. 60
10.35 a.m. Gassed	10.35 a.m. Gassed
11.00 a.m. Continuous oxygen by mask, 1 liter per minute	11.00 a.m. Continuous oxygen by mask, 1 liter per minute
11.40 a.m. Hb. 79 (100 per cent)	11.50 a.m. Hb. 70 (117 per cent)
2.20 p.m. Hb. 85 (106 per cent)	2.40 p.m. Hb. 84 (140 per cent)
2.45 p.m. Continuous oxygen by mask, 3 liters per minute	2.45 p.m. Continous oxygen by mask, 3 liters per minute
4.25 p.m. Hb. 105 (131 per cent)	3.45 p.m. Hb. 105 (175 per cent). Heart puncture. Venous blood 55 per cent saturated. Animal died on table. L: H 8.0
4.45 p.m. Heart puncture. Arterial blood 93 per cent saturated. Venous blood 45 per cent saturated	
5.45 p.m. Hb. 110 (137 per cent)	
5.55 p.m. Died. L: H 8.2	

GOAT 4406 (CONTROL)	GOAT 4542 (CONTROL)
9.45 a.m. Hb. 42	9.50 a.m. Hb. 74
10.35 a.m. Gassed	10.35 a.m. Gassed
12.10 p.m. Hb. 48 (114 per cent)	12.20 p.m. Hb. 76 (103 per cent)
2.30 p.m. Hb. 60 (143 per cent)	2.50 p.m. Hb. 88 (119 per cent)
3.00 p.m. Died. L: H 6.0	5.30 p.m. Hb. 96 (130 per cent). Found dead next morning. L: H 8.3

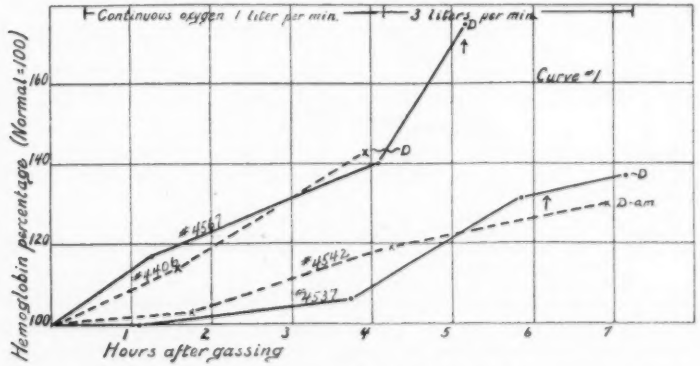

Fig. 1. Showing the concentration of the blood after gassing. Solid line: animals receiving extra oxygen. Arrows indicate time of heart puncture. *No. 4567*, venous blood 55 per cent saturated. *No. 4537*, arterial blood 93 per cent saturated; venous blood 45 per cent saturated. Dotted line: control animals.

TABLE 2

Chlorpicrin 1/8500 for 30 minutes (9.30 to 10.00 a.m.), August 21, 1918

GOAT 4526	GOAT 4446 (CONTROL)
5.35 p.m. (8/19). Hb. 53	5.30 p.m. (8/19). Hb. 72
10.00 a.m. Gassed	10.00 a.m. Gassed
10.15 a.m. Continuous oxygen by mask, 1½ liters per minute	12.15 p.m. Hb. 96 (133 per cent)
12.00 m. Hb. 67 (126 per cent)	1.45 p.m. Hb. 118 (164 per cent)
12.15 p.m. Continuous oxygen by mask, 4 liters per minute	2.00 p.m. Died.
2.10 p.m. Hb. 70 (132 per cent)	
3.20 p.m. Hb. 73 (138 per cent)	
3.50 p.m. Hb. 76 (143 per cent). Heart puncture. Arterial blood 90 per cent saturated. Venous blood (obtained only after struggling) 20 per cent saturated	
4.15 p.m. Oxygen stopped	
5.30 p.m. Hb. 76 (143 per cent)	
6.05 p.m. Died. L: H 7.7	

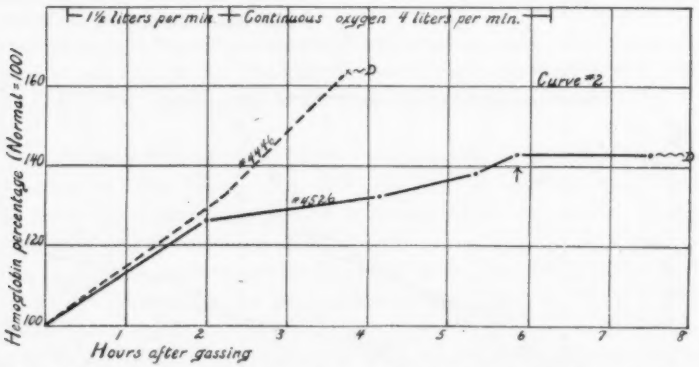

Fig. 2. Showing the concentration of the blood after gassing. Solid line: animals receiving extra oxygen. Arrow indicates time of heart puncture. *No. 4526*, arterial blood 90 per cent saturated. Dotted line: control animals.

TABLE 3

Chlorpicrin 1/8500 for 25 minutes (9.16 to 9.41 a.m.), August 23, 1918

GOAT 4577	GOAT 4631
5.50 p.m. (8/22). Hb. 60	5.45 p.m. (8/22). Hb. 58
9.41 a.m. Gassed	9 41 a.m. Gassed
9.55 a.m. Continuous oxygen by mask, 3 liters per minute	9.55 a.m. Continuous oxygen by mask, 3 liters per minute
10.23 a.m. Hb. 70 (117 per cent)	10.10 a.m. Hb. 54 (93 per cent)
12.12 p.m. Hb. 81 (135 per cent)	12.00 m. Hb. 58 (100 per cent)
2.35 p.m. Hb. 95 (158 per cent). Heart puncture. Both samples obtained only after struggling. Arterial blood 58 per cent saturated. Venous blood 4 per cent saturated	2.50 p.m. Hb. 61 (105 per cent)
	5.00 p.m. Hb. 67 (116 per cent)
	5.55 p.m. Hb. 69 (119 per cent)
	6.20 p.m. Heart puncture. Arterial blood 95 per cent saturated. Venous blood about 50 to 60 per cent
2.40 p.m. Died on table. L: H 7.4	6.20 p.m. Hb. 74 (128 per cent). Died on table. L: H 5.7

GOAT 4490 (CONTROL)	GOAT 4457 (CONTROL)
9.41 a.m. Gassed	9.41 a.m. Gassed
10.43 a.m. Hb. 85	10.54 a.m. Hb. 94
12.30 p.m. Hb. 100 (118 per cent)	12.20 p.m. Hb. 96 (102 per cent)
2.15 p.m. Hb. 124 (146 per cent)	3.15 p.m. Hb. 106 (113 per cent)
2.20 p.m. Died. L: H 6.7	6.30 p.m. Hb. 124 (132 per cent). Found dead next morning. L: H 7.1

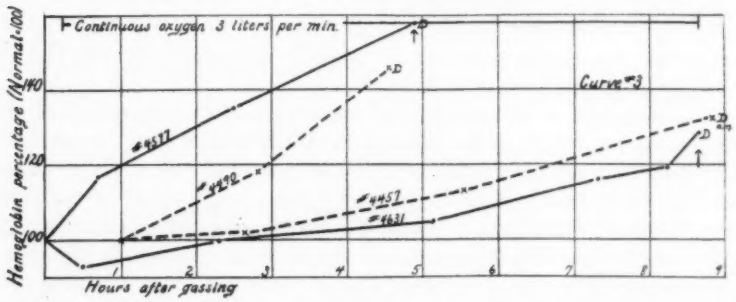

Fig. 3. Showing the concentration of the blood after gassing. Solid line: animals receiving extra oxygen. Arrows indicate time of heart puncture. *No. 4577*, both blood samples obtained only after struggling. *No. 4631*, arterial blood 95 per cent saturated; venous blood 50 per cent saturated.

centrated as rapidly and to as great an extent as that of the control animals.

In order to demonstrate the efficiency of the oxygen administration, samples of blood were taken from both sides of the heart at suitable intervals and analyzed for oxygen. In most of the experiments the venous and arterial blood samples were obtained from the heart without difficulty and contained hemoglobin which was normally saturated with oxygen. With the increased concentration of the hemoglobin the oxygen content of the blood was even above normal.

Occasionally the blood was obtained only after considerable struggling on the part of the animal so that the reduced oxygen content of such bloods was to be expected. These observations are reported here merely to make the experimental record complete, as obviously the low oxygen content of such bloods is without bearing on the present problem.

These experiments demonstrate that, by breathing oxygen-rich atmospheres, oxygen may be absorbed through the damaged and edematous lungs in quantities sufficient to maintain a practically normal level of oxygen in the arterial blood. The high saturation of the hemoglobin of venous blood with oxygen would seem to prove that the blood flow is sufficiently rapid to normally oxygenate the tissues. Nevertheless, in spite of the normal oxygenation of the tissues in the animals receiving oxygen, the blood concentrated as rapidly and to as great an extent as in the control animals. The conclusion therefore seems justifiable that the lack of oxygen in the tissues and consequent imbibition of water is not an important factor in causing the concentration of the blood in animals developing edema after being gassed with lung irritants.

An indication of the severity of the lung edema was obtained by comparing the weight of the lung to the weight of the heart at autopsy. The high lung to heart ratios obtained in practically all of the animals studied show that a severe grade of edema had already developed. The extent of the edema as indicated by this method was as great in the animals receiving oxygen as in the controls. Although the data are necessarily few, it is apparent that the efficient oxygenation of the lung tissue in the animals receiving oxygen failed to diminish the tendency for the development of the edema of the lungs.

With the enormous accumulation of fluid in the edematous lungs and the loss of water from the blood running roughly parallel, it is a tempting study to estimate even in a rough way the possible water

interchange. An attempt has been made with data which are more or less incomplete and with calculations involving gross errors but the relations are so striking that they are presented here (table 4). In the table are recorded data and calculations from animals in which the hemoglobin was not determined immediately before death but is estimated from the curve obtained from the various determinations. These estimated values are quite similar to average values obtained at death on other animals.

TABLE 4

Comparison of calculated amounts of fluid lost from blood and extra fluid in the lungs in gassed animals

(1) GOAT NUMBER	(2) DATE	(3) WEIGHT ANIMAL	(4) WEIGHT HEART AT DEATH	(5) WEIGHT LUNG AT DEATH	(6) L: H	(7) WEIGHT NORMAL LUNG	(8) LAST HEMOGLOBIN DETERMINATION	(9) HEMOGLOBIN AT DEATH	(10) NORMAL BLOOD VOLUME	(11) BLOOD VOLUME AT DEATH	(12) FLUID LOST FROM BLOOD AT DEATH	(13) EXTRA FLUID IN LUNG
		kgm.	*gram*	*gram*		*gram*	*per cent*	*per cent*	*cc.*	*cc.*	*cc.*	*cc.*
3602	6/13	31.8	157	1472	9.4	377	163	200	1750	875	875	1095
3638	6/12	29.5	126	623	4.9	302	125	140	1625	1160	465	321
3787	6/20	16.8	74	566	7.6	178	128	145	925	638	287	388
4098	7/ 4	35.0	195	1103	5.7	468	114	140	1925	1375	550	635
4406	8/19	15.5	85	510	6.0	204	143	150	850	567	283	306
3920	7/ 4	37.8	200	1700	8.5	480	180	*	2080	1155	925	1220
3600	6/12	25.5	130	849	6.5	312	154	*	1400	910	490	537
4047	7/12	33.6	210	1556	7.4	504	130	*	1850	1420	430	1052

* Time of death not known. Calculations made using last hemoglobin determinations (column 8).

Column 6 = column 5 ÷ column 4.

Column 7 = column 4 × 2.4.

Columns 8 and 9. Values calculated using the normal as 100 per cent.

Column 9 = extrapolation to time of death.

Column 10 = column 3 × 0.055 × 1000 (Boycott and Damant, Journ. Physiol., 1907–8, xiv, 36).

Column 11 = column 10 ÷ column 9.

Column 12 = column 10 − column 11.

Column 13 = column 5 − column 7.

Examining the last two columns of the table it is evident that in only one instance the amount of extra fluid in the lung is less than the calculated loss of fluid from the blood. In some instances the extra fluid in the lung is much greater than that lost by the blood. Little or no water is drunk by goats in this condition and the volume of urine

excreted is small, so that the external factors do not confuse the picture. Even with the relatively large errors of calculation involved, the conclusion seems justified that the loss of fluid by the blood can be accounted for by the excess of liquid in the edematous lung.

The evidence suggests that the muscles, etc., do not imbibe water and cause the concentration of the blood. In fact it would appear that water may be drawn from some tissues to make up part of the volume of liquid in the lung. We are thus finally led back to our original point of view that the development of the edema of the lungs and the concentration of the blood are interrelated and are the important factors in the pathological condition studied. With this fact established it is justifiable to conclude that the development of the edema of the lungs is the primary factor in the condition and that the development of the edema causes the concentration of the blood.

SUMMARY

The continuous administration of oxygen to goats gassed with chlorpicrin did not inhibit the concentration of the blood.

The percentage saturation of the hemoglobin with oxygen was normal even after a considerable concentration of the blood had occurred.

The concentration of the blood is not caused by the imbibition of water by the tissues as the result of oxygen want.

The loss of water from the blood is therefore due to the development of the edema of the lungs.

We wish to thank Mr. J. Barcroft and the staff of the Physiological Laboratories of the R. E. Experimental Station, Porton, England, for the many kindnesses extended to us throughout the course of our investigations there.

THE EFFECT OF ADRENALIN, DESICCATED THYROID AND CERTAIN INORGANIC SALTS ON CATALASE PRODUCTION

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Dahm and Steck (1) found that the ingestion of urea and of sodium chloride increased oxidation. This observation has been repeated and confirmed by Tangl (2) on curarized animals with their kidneys removed. Raeder (3) also found that saline injections especially if hypertonic increased the respiratory exchange. Loewy (4) observed that the ingestion of small amounts of water (100 cc.) produced no change in oxygen consumption, while Speck (5) found that drinking large quantities (1250 cc.) did. According to Lusk (6) water, sodium chloride and urea have no effect on the respiratory exchange. Magnus-Levy (7) observed an increased carbon dioxide output in a man fed thyroid extract and an increased oxygen intake in cases of exophthalmic goiter. It is recognized that tetrahydro-β-naphthylamin increases oxidation in the body and decreases heat elimination. Grafe (8) has shown that the administration of ammonium carbonate, ammonium chloride and sodium carbonate increases oxidation.

We (9) had found that whatever increased oxidation in the body, the ingestion of food, for example, produced an increase in catalase by stimulating the alimentary glands, particularly the liver, to an increased output of this enzyme, and that whatever decreased oxidation, narcotics, for example, produced a decrease in catalase by direct destruction and by decreasing its output from the liver. The object of the present investigation was to determine if adrenalin, tetrahydro-β-naphthylamin, desiccated thyroid, water, sodium chloride, ammonium chloride, sodium carbonate, ferric chloride, sodium citrate, ammonium carbonate, urea, triacetin and saccharin would or would not produce an increase in catalase. The amounts of the substances used will be given in the description of the individual experiments. The animals used were dogs and rabbits. After etherizing these animals

and opening the abdominal wall, each of the substances was introduced into the upper part of the intestine. The catalase in 0.5 cc. of blood was determined before, as well as at fixed intervals after the introduction of the materials. The determinations were made by adding 0.5 cc. of blood to hydrogen peroxide in a bottle at approximately 22°C. and the amount of gas liberated in 10 minutes was taken as a measure of the catalase content of the 0.5 cc. of blood.

In figure 1 are shown the effects of the introduction into the intestines of rabbits of urea, sodium chloride, water, triacetin and glycerine. The amounts of the substances used are indicated on the chart. Seventy-five cubic centimeters of water were used in dissolving the different substances. The figures along the ordinate (0 to 780) indicate amounts of catalase measured in cubic centimeters of oxygen and those along the abscissae time in minutes.

It may be seen that 2 grams of urea, 1 gram of sodium chloride and 15 cc. of water per kilo produced no increase in catalase in keeping with Lusk's observation that amounts of these substances as small as these produced no increase in oxidation, while 10 grams of urea, 10 grams of sodium chloride and 150 cc. of water per kilo did produce an increase in catalase in keeping with the observations of Dahm and Steck, Tangl, Speck and Raeder, that large amounts of these substances produce an increase in oxidation.

It may also be seen that triacetin is more effective in producing an increase in catalase than glycerine, this result being attributed to the presence of the acetic acid radicle in the glycerine molecule.

The second part of the paper is concerned with determining the mode of action of the substances already mentioned as well as several other substances in producing an increase in catalase. The animals used were dogs and the method for determining catalase was the same as that already described. The substances were dissolved in 200 cc. of distilled water and were introduced at body temperature into the upper part of the small intestine. The catalase content of 0.5 cc. of blood taken from the liver, portal and jugular veins was determined before as well as at certain fixed intervals after the introduction of the materials. In figures 2, 3 and 4 the continuous line curves were constructed from data obtained from the blood of the liver, the discontinuous line curves from the blood of the portal, and the dotted line curves from that of the jugular vein.

In figure 2 it may be seen, as we have found before, that the introduction of glycocoll produced an increase in catalase as is indicated by

the increase in the amount of oxygen liberated by the blood. It may be seen further that this increase is greater, particularly during the first fifteen minutes, in the blood of the liver than in the blood of the jugular or portal veins.

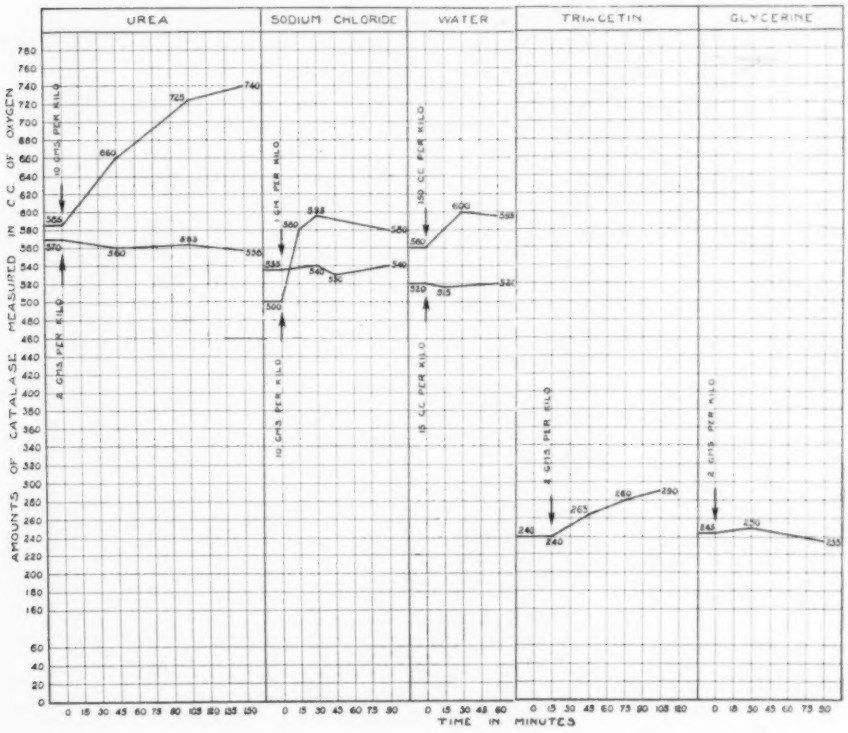

Fig. 1. The effect of the introduction into the intestines of rabbits of the substances named in the chart on blood catalase.

Under urea it may be seen as with glycocoll that this substance increases the catalase of the blood of the liver more rapidly than that of the portal and jugular veins. Under glycerine and triacetin it may be seen that while 5 grams per kilo of glycerine produced no increase in catalase, a similar amount of triacetin produced an increase thus showing that triacetin is more effective in this respect than

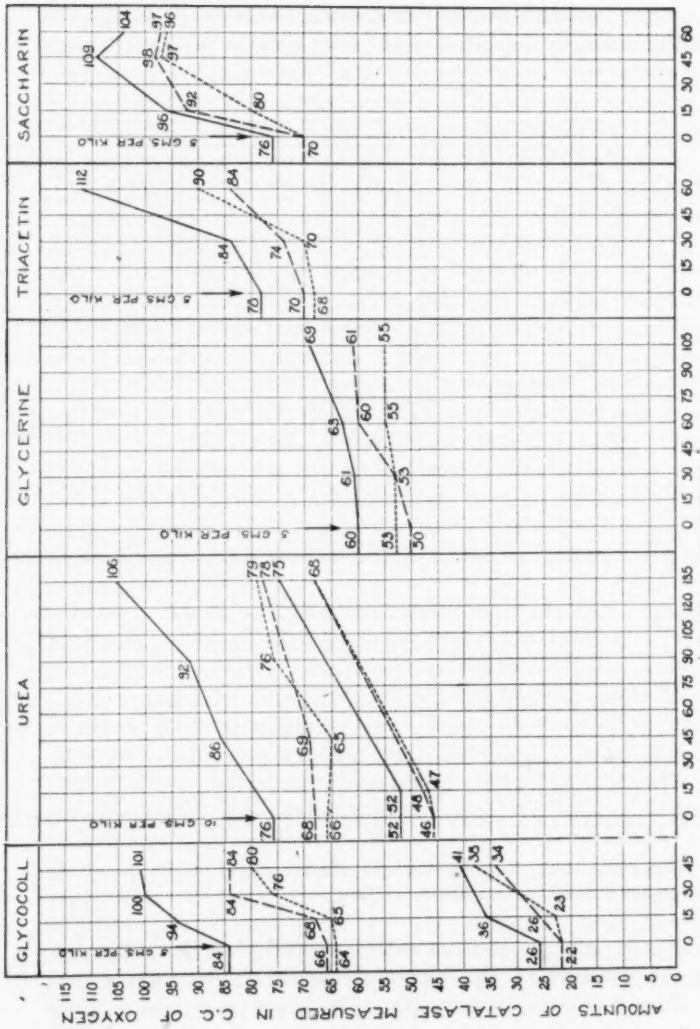

Fig. 2. The effect of the introduction into the intestines of dogs of the substances named in the chart on blood catalase. The continuous line curves were constructed from data obtained from the blood of the liver; the discontinuous ones from the blood of the portal and the dotted line curves from the blood of the jugular vein.

glycerine. It may also be seen that 5 grams per kilo of saccharin produced an increase in catalase. It (10) had been found that 5 grams of sugar would not produce so large an increase in catalase as is here shown to be produced by this amount of saccharin, hence so far as

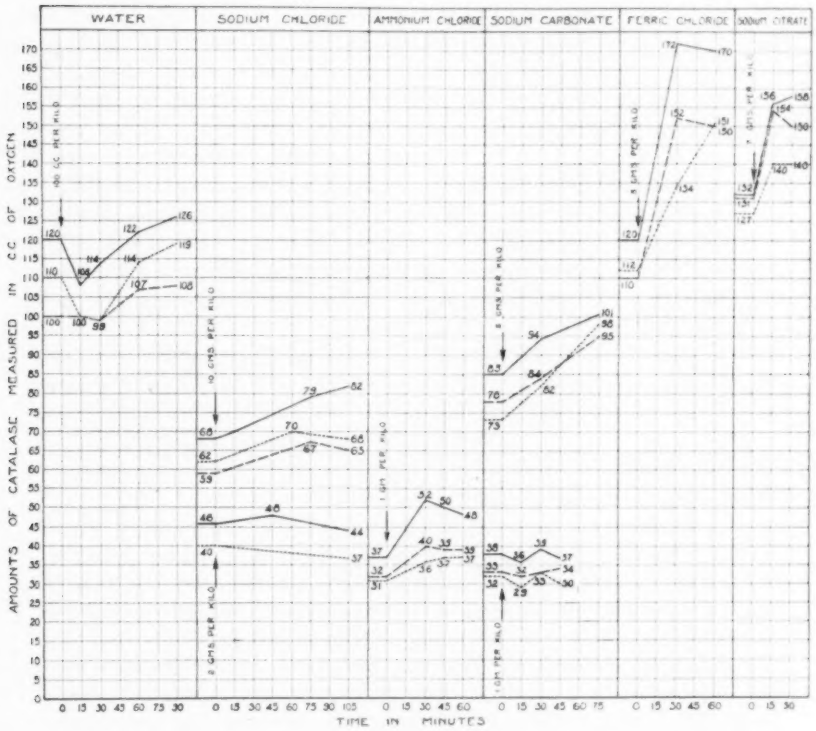

Fig. 3. The effect of the introduction into the intestines of dogs of the substances named in the chart on blood catalase. The continuous line curves were constructed from data obtained from the blood of the liver; the discontinuous ones from the blood of the portal and the dotted line curves from the blood of the jugular vein.

catalase production is concerned saccharin is more effective than sugar. It would seem that in addition to being a sweetening agent, saccharin, although not oxidized itself to give rise to energy, as is the case with sugar, serves to stimulate the alimentary glands to an in-

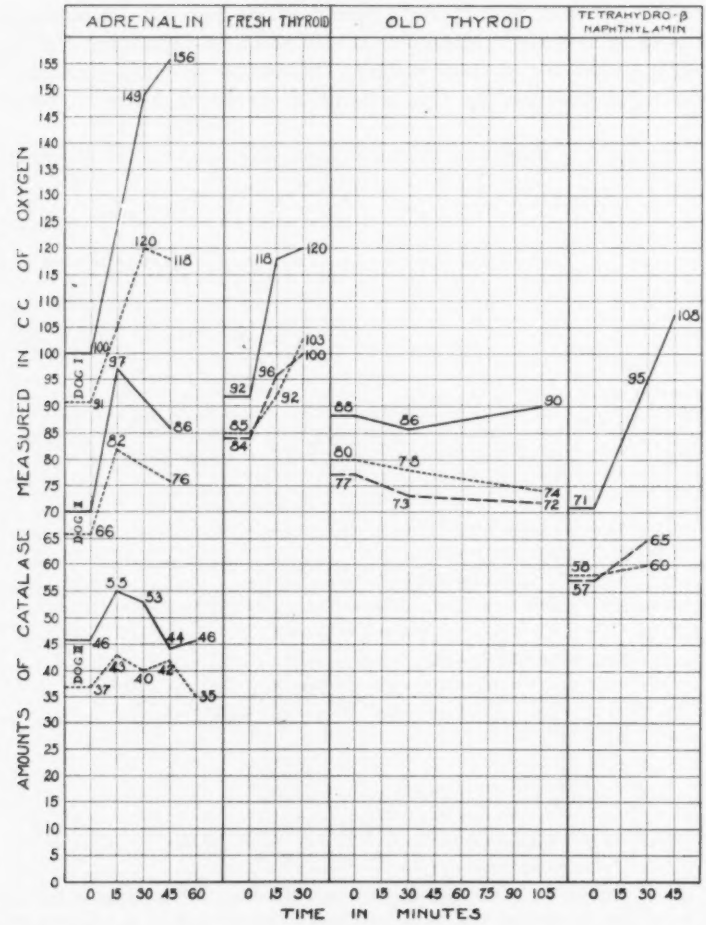

Fig. 4. The effect of the introduction into the intestines of dogs of the substances named in the chart on blood catalase. The continuous line curves were constructed from data obtained from the blood of the liver; the discontinuous ones from the blood of the portal and the dotted line curves from the blood of the jugular vein.

creased output of catalase thus facilitating the oxidation of the other food materials.

In figure 3 it may be seen that the introduction of water, sodium chloride, ammonium chloride, sodium carbonate, ferric chloride and sodium citrate into the alimentary tract of dogs produced an increase in the catalase of the blood and that this increase was brought about by stimulating the alimentary glands, particularly the liver, to an increased output of this enzyme. The fact that these salts produce an increase in catalase and hence in oxidation may in some measure account for the beneficial effect of certain mineral waters which contain these substances in conspicuous amounts.

In figure 4 is shown the effect of adrenalin, freshly prepared and old thyroid and tetrahydro-β-naphthylamin on catalase production. Three cubic centimeters of a 1: 1000 solution of adrenalin chloride were introduced into the portal vein in dog 3 and 5 cc. in dog 2. In these two dogs the solutions were injected as quickly as could be conveniently done while in dog 1, 10 cc. of a 1: 1000 adrenalin chloride solution diluted to 50 cc. were injected at a rate of about 1 cc. per minute, thus requiring approximately 30 minutes for the injection. It may be seen that the adrenalin increased the catalase of the blood by stimulating the liver to an increased output of this enzyme.

As a result of the work of Blum (11), Vosburgh and Richards (12), Dreyer (13), Oliver and Schaefer (14), Cannon and de la Paz (15) it is now believed that during combat the adrenals are stimulated to an increased output of adrenalin and that this produces a constriction of the small blood vessels of the abdominal viscera, thus increasing the blood supply to the heart, skeletal muscles and nervous system; that it hastens the coagulation of the blood and increases the output of sugar from the liver. It is evident that the result of diverting the blood from the abdominal viscera into the heart, skeletal muscles and nervous system during combat is to render conditions more favorable for the increased action of these organs; that the hastening of the coagulation is to stop more quickly the bleeding from any superficial wound that may be inflicted, and that the flushing of the blood with sugar is to insure a plentiful supply of oxidizable material to the muscles. While the preceding hypothesis explains certain phases of adaptation of the organism for combat, it does not explain how the increased oxidation is brought about which gives rise to the energy for the fight. We had already found that the stimulation of the splanchnic nerves to the liver produced an increased output of catalase

from this organ, and had suggested that the increased oxidation during combat might be due to this increase in catalase. The fact that adrenalin stimulates the liver to an increased output of catalase suggests that this result may occur during combat when there is an increased output of adrenalin into the blood.

It may be seen in the figure that the introduction into the alimentary tract of freshly prepared thyroid produced an increase in catalase while old thyroid did not. The old thyroid was a Parke-Davis preparation which had been standing in the laboratory for four years, while the fresh thyroid was material recently purchased from this same company. The old material was fed to cats and found to have lost its virtue while the new material produced the characteristic effects, loss of weight, etc. The amount of the thyroid introduced into the intestines of the dogs was 1 gram per kilo dissolved in 200 cc. of water.

We had already found that the feeding of thyroid to cats increased very greatly the catalase of the blood. The experiments described in this paper on the introduction of thyroid into the alimentary tract of dogs suggest that the increase in the catalase of the blood of animals fed with thyroid is due to the stimulation of the liver to an increased output of this enzyme. Winternitz (16) found that "the removal of the thyroid gland caused a drop in the catalase activity of the blood which was compensated if thyroid were fed" and that in hyperthyreosis the catalase of the blood tends to increase while in hypothyreosis it assumes a lower level than normal. Becht (17), on the other hand, claims that thyroid feeding decreases the catalase of the blood. It should be mentioned also in this connection that Becht holds that narcotics slightly increase the catalase content of the blood, while we found that they produce a great decrease both in vivo and in vitro.[1]

Under tetrahydro-β-naphthylamin it may be seen that the introduction into the intestine of 0.8 gram per kilo of this substance dissolved in 200 cc. of water stimulated the liver very greatly to an increased output of catalase which is offered in explanation of the increased oxidation produced by this substance.

[1] Owing to our proximity it has been suggested and even urged that Doctor Becht and myself carry out some joint experiments in an attempt to clear up the differences in our results. I am sorry to say that Doctor Becht seems to be unwilling to carry out such experiments.

SUMMARY

The introduction into the alimentary tract of relatively small amounts of water (15 cc.), of sodium chloride (1 gm.), and of urea (2 gms.) per kilo, produces no increase in catalase in keeping with Lusk's observation that small amounts of these substances produce no increase in oxidation, while the introduction of large amounts of these substances, 1500 cc. of water, 10 grams of urea per kilo and 10 grams of sodium chloride per kilo do produce an increase in catalase in keeping with the observations of Dahm and Steck, Tangl, Speck and Raeder, that large amounts of these substances produce an increase in oxidation.

The injection of adrenalin into the portal vein stimulates the liver to an increased output of catalase. This fact suggests that the increased amount of adrenalin thrown into the circulation during combat may stimulate the liver to an increased output of catalase, and in this way aid in bringing about the increase in oxidation occurring during combat.

Desiccated thyroid when introduced into the alimentary tract stimulates the liver to an increased output of catalase. This observation suggests that the increase in the catalase of the blood which may be responsible for the increase in the respiratory exchange of an animal when fed with thyroid or in exophthalmic goiter is probably due to the stimulation of the liver to an increased output of catalase.

BIBLIOGRAPHY

(1) Dahm and Steck: Physiol. Gesellsch. zu Berlin Verhandl., 1909, xxxiv.
(2) Tangl: Biochem. Zeitschr., 1911, xxxiv, 1.
(3) Raeder: Biochem. Zeitschr., 1915, lxix, 257.
(4) Loewy: Arch. f. d. gesammt. Physiol. d. Menschen, 1888, xliii, 515.
(5) Speck: Policlinico II, 1892, xlii.
(6) Lusk: Journ. Biol. Chem., 1912, xiii, 27.
(7) Magnus-Levy: Berl. klin. Wochenschr., 1895, xxx, 650.
(8) Grafe: Deutsch. Arch. f. klin. Med., 1915, cxviii, 1.
(9) Burge: This Journal, 1919, xlviii, 133.
(10) Burge: Med. Rec., December, 1918.
(11) Blum: Arch. gesammt. Physiol., 1902, xc, 617.
(12) Vosburgh and Richards: This Journal, 1903, ix, 29.
(13) Dreyer: This Journal, 1899, ii, 283.
(14) Oliver and Schaefer: Journ. Physiol., 1895, xviii, 230.
(15) Cannon and De La Paz: This Journal, 1911, xxviii, 64.
(16) Winternitz: Arch. Int. Med., 1911, vii, 625.
(17) Becht: This Journal, 1919, xlviii, 171.

A NOTE ON THE QUESTION OF THE SECRETORY FUNCTION OF THE SYMPATHETIC INNERVATION TO THE THYROID GLAND

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Considerable interest has attached itself to the question of the innervation of the thyroid gland, and as yet the conflicting evidence presented by different experimenters does not permit the drawing of definite conclusions. Asher and Flack (1), by stimulating the laryngeal fibers from the vagus, found the blood pressure response to adrenalin injection to be greatly increased. Asher (2) later decided that this increased effectiveness of adrenalin was due to the sensitizing action of the thyroid secretion on sympathetic endings following stimulation of the nerve fibers to the gland. Cannon and Cattell (3) demonstrated an electrical variation in the gland, which they interpreted as evidence of secretory activity following stimulation of the cervical sympathetic fibers, while stimulation of laryngeal fibers from the vagus was ineffective. Levy (4) observed an increased blood pressure response to adrenalin after stimulation of cervical sympathetic and after adrenalin injection, which he accepted as evidence of sensitization of sympathetic endings by the thyroid secretion following stimulation. Cannon, Ringer and Fitz (5), by fusion of the central end of the phrenic nerve to the peripheral portion of the cut cervical sympathetic trunk, obtained symptoms of sympathetic stimulation in the eye and in the ear vessels of rabbits, and also symptoms similar to those of hyperthyroidism, such as rapid pulse, increased metabolism and hyperexcitability. These latter symptoms disappeared on removal of the thyroid on the side of the nerve suture, so the evidence was taken to indicate thyroid stimulation by the phrenic impulses.

On the other hand Troell (6) failed to obtain any evidence of thyroid stimulation, either symptomatically or microscopically, on suturing the phrenic to the cervical sympathetic. Burget (7), and Marine,

Rogoff and Stewart (8) also reported negative results by this same method. Manley and Marine (9) and others, have reported that thyroid transplants survive and function in various parts of the body, regardless of nerve supply, so that secretory nerves to the gland are at least not essential to its activity.

From the above contradictory evidence presented it is evident that any new contribution should be welcome. The object of the experiments described in this paper was to determine whether the thyroid could be stimulated to greater activity through repeated injections of cocaine, the basis of the use of cocaine being the work of Froelich and Loewi (10), in which they demonstrate quite conclusively that cocaine sensitizes sympathetic nerve endings to the action of adrenalin and to sympathetic stimulation.

EXPERIMENTAL

The subcutaneous injection of 35 to 50 mgm. cocaine hydrochloride per kilo body weight, or of 10 mgm. cocaine hydrochloride accompanied by 1 mgm. adrenalin per kilo body weight, calls forth in the rabbit symptoms of undoubted sympathetic stimulation, such as maximal dilatation of the pupils, slight exophthalmos, constriction of the ear vessels leaving the ears cold to the touch, relaxation and loss of tone in the intestines, and erection of the hair over the body. That it is either a sensitization of the sympathetic endings to the normal flow of impulses over these nerves, or else an increased central stimulation, can be shown by cutting one sympathetic nerve in the neck and then noting the effect of cocaine on the pupils and ear vessels. Effects of the drug were seen only on the side having the intact nerve supply. Further, on frightening the animal, as by dropping from a distance of a foot or so onto the observation table, a slight dilatation of the pupil on the side of the intact nerve occurs within one-half second and disappears within one second after cessation of the frightening, thus indicating that the results obtained are not due to a sudden outpouring of epinephrin, but far more likely due to impulses traversing the sympathetic nerves.

Having established to my satisfaction, then, that cocaine acts in the normal rabbit by increasing the effectiveness of the normally occurring sympathetic impulses, I next tried to determine the effects on the thyroid gland as evidenced by histological changes in its structure. Specimens of the gland were removed aseptically before and after the cocaine treatment and examined for changes in their microscopic

appearance. Ordinary aseptic precautions were observed in the injections. Since all references in the literature indicate that cocaine is only slowly eliminated, (Wiechowski (11), Grode (12) the treatment was begun with daily injections of sufficient strength to produce a maximal mydriasis of the pupils lasting about 30 minutes, that is, about 10 mgm. of cocaine per kilo body weight. However, since the effects seemed to be over within an hour, the injections were increased to 3 or 4 a day. In one series of five rabbits thus treated for 8 days there was observed no change whatever in the microscopic appearance of the thyroid glands. A second series of three rabbits was run, each animal receiving 5 to 10 injections a day for 11 days, and again no changes in the glands were observed. Neither did the rate of growth or general appearance and behavior of the animals indicate any lasting results from the use of the drug.

Since the strength or effectiveness of the impulses over the sympathetic fibers should have been greatly augmented by the continued use of cocaine producing symptoms of thyroid hyperactivity and morphological changes in the gland, the evidence from these experiments contributes to the indication of a lack of secretory function of the sympathetic fibers to the thyroid gland.

Note. The experimental part of this work was carried on in the Laboratory of Pharmacology of the University of Chicago at the suggestion, and under the direction, of Dr. A. L. Tatum, for whose aid and suggestions I desire to express my sincere thanks.

BIBLIOGRAPHY

(1) Asher, Leon and Flack: Zeitschr. f. Biol., 1910, lv, 83.
(2) Asher and Leon: Deutsch. Med. Wochenschr., 1916, xxxiv, 1.
(3) Cannon, and Cattell: This Journal, 1916, xli, 58.
(4) Levy: This Journal, 1916, xli, 492.
(5) Cannon, Binger and Fitz: This Journal, 1914, xxxvi, 363.
(6) Troell: Arch. Int. Med., 1916, xvii, 382.
(7) Burget: This Journal, 1917, xliv, 492.
(8) Marine, Rogoff and Stewart: This Journal, 1918, xlv, 268.
(9) Manley and Marine: Proc. Soc. Exper. Biol. and Med., 1914, xii, 202.
(10) Froelich and Loewi: Arch. f. Exper. Path., 1910, lxii, 158.
(11) Wiechowski: Arch. f. Exper. Path. u. Pharm., 1901, xlvi, 155.
(12) Grode: Arch. f. Exper. Path., 1912, lxvii, 172.